MARINE ECOLOGY - MACKENZIE DELTA AND TUKTOYAKTUK PENINSULA

A SERIES OF FOUR REPORTS ON THE MARINE ECOLOGY OF THE MACKENZIE DELTA AND TUKTOYAKTUK PENINSULA REGION

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for the

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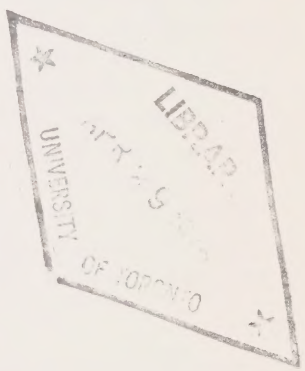

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The data for these reports were obtained as a result of investigations carried out under the Environmental-Social Program, Northern Pipelines, of the Task Force on Northern Oil Development, Government of Canada. While the studies and investigations were initiated to provide information necessary for the assessment of pipeline proposals, the knowledge gained is also useful in planning and assessing other development projects.

RÉSUMÉ EN FRANCAIS

Partie I

Les responsables d'une étude préliminaire de la biodégradation du pétrole dans les eaux marines de l'Arctique se sont efforcés de quantifier et d'isoler des bactéries oléodégradantes dans les lacs des Esquimaux, T.N.-O. Le procédé employé n'a pas permis de déterminer leur abondance dans ces lacs. On a cependant réussi à isoler des bactéries oléodégradantes dans les eaux marines au moyen d'une méthode d'enrichissement. On a constaté, en laboratoire, que des cultures mixtes obtenues grâce à cette méthode dégradaient dans une certaine mesure trois bruts différents, dont un de Norman Wells à une température d'à peine 2°C. Les essais n'ont pas porté sur des températures inférieures. Selon une expérience préliminaire, il semblerait que les concentrations de phosphore et d'azote des eaux marines ne constituent pas un facteur limitatif de la dégradation à basse température, mais des études complémentaires sont nécessaires dans ce domaine.

Partie II

On ne sait que peu de choses des conséquences biologiques à court ou à long terme de la pollution par les hydrocarbures sur les populations d'organismes marins dans l'Arctique. Se basant sur les recherches très poussées effectuées dans les régions tempérées, on avait prédit que ses effets seraient particulièrement graves sur les écosystèmes de l'Arctique, en particulier du point de vue de leur rythme de récupération après qu'ils aient subi des dommages. La présente étude passe en revue les nombreux textes traitant des effets du pétrole sur les invertébrés marins pris dans leur ensemble, afin de préciser l'ampleur potentielle et la nature des risques que présente le pétrole pour la vie marine et de mieux situer l'étude dans son contexte.

Cette étude a été conçue pour rassembler des renseignements sur les interactions nocives possibles entre certains des invertébrés marins dominant dans les eaux côtières peu profondes voisines du Delta du MacKenzie et le pétrole brut de l'Arctique. Deux échantillons de ce dernier ont été utilisées, l'un de Norman Wells et l'autre du puits d'Atkinson, sur la péninsule Tuktoyaktuk adjacente à la zone d'étude. A des fins de comparaison, on a également utilisé pour certaines expériences un échantillon de brut vénézuélien obtenu récemment (pétrole qui a fait l'objet d'études considérables en ce qui concerne ses effets biologiques).

Le brut peut causer des dommages directs aux organismes de deux manières distinctes. Certains de ses composants peuvent endommager les tissus par une action chimique sur la structure ou le fonctionnement des cellules. D'un autre côté, du fait de sa viscosité éleyée, le pétrole peut endommager physiquement les organismes en mettant obstacle à leurs facultés de locomotion, d'alimentation, de respiration, etc. Les travaux exposés ici portent sur ces deux types de dommages.

Un certain nombre d'espèces ont fait l'objet d'une étude de la toxicité à court terme conçue pour déterminer les dommages chimiques provoqués par certains composants du pétrole solubles dans l'eau de mer. Les formes adultes de crustacés benthiques examinées (un amphipode et un cumacé) se sont révélées plutôt tolérantes à de fortes concentrations de pétrole dans l'eau de mer. Un bibalve benthique s'est également révélé très tolérant, bien qu'il manifeste initialement une réaction de fuite dont l'amplitude et la durée sont directement fonction de la concentration en pétrole. Un organisme benthique tuniqué s'est révélé très sensible au pétrole et a succombé à des concentrations modérées. Les recherches n'ont porté que sur une espèce planctonique. Cette petite méduse, localement abondante, s'est révélée très sensible à des concentrations plutôt faibles de pétrole dans l'eau de mer et, de ce fait, la moins tolérante de toutes les espèces examinées.

Il est important de noter que ces résultats sont basés sur des études de l'effet létal à court terme et que même les espèces classées ici comme tolérantes au pétrole brut peuvent très bien souffrir et finalement succomber si la période d'exposition est plus longue. Pour cette raison, il est nécessaire de définir des critères physiologiques plus sensibles afin de déterminer les effets délétères sub-létaux. L'activité métabolique est un indicateur sensible de l'état physiologique général d'un organisme. Les chercheurs ont étudié les effets d'une exposition à court terme aux composants du brut de l'Arctique solubles dans l'eau de mer sur le métabolisme de l'amphipose Onisimus affinis. Le brut d'Atkinson Point et celui de Norman Wells n'ont pas eu d'effets significatifs sur la respiration à des concentrations faibles ou modérées. Celui d'Atkinson Point augmente la respiration de 25% à une concentration de 1,000 millionièmes, et de 30% à 5,000 millionièmes. Celui de Norman Wells l'augmente respectivement de 25% et de 38%. Selon certaines indications, avec des concentrations encore plus élevées de ces deux bruts, cette activité commence à diminuer.

Il semble que des formes benthiques adultes telles que Onisimus ne subissent pas d'effets toxiques directs dus aux composants du pétrole brut solubles dans l'eau de mer aux concentrations les plus courantes dans le milieu naturel. Les effets physiques résultant d'un contact direct avec des quantités massives de pétrole seraient sans doute beaucoup plus dommageables. Cela est particulièrement vrai pour les espèces qui, telles Onisimus, bien que normalement benthiques, se concentrent en grands nombres au voisinage de la face interne de la glace au cours de l'hiver. C'est en effet à cet endroit que s'accumuleront probablement des poches de pétrole. Une série d'expériences ont été effectuées pour déterminer les facultés de récupération d'Onisimus replacé dans l'eau de mer propre après une brève immersion dans plusieurs bruts. La mortalité augmente progressivement avec le temps et "la vitalité" des animaux (mesurée quantitativement par un indice d'activité) diminue progressivement. Quatre semaines après l'exposition, moins de 20% des animaux sont capables de nager, la plupart du temps d'une manière erratique et anormale. Très peu de ces animaux, pour ne pas dire aucun, ont semblé s'en remettre complètement.

Étant donné le fait que le contact physique avec du pétrole brut peut présenter les plus grands risques potentiels pour certaines espèces, il est nécessaire de déterminer la réponse éthologique des organismes marins aux masses de pétrole présentes dans leur voisinage immédiat. Afin de permettre les comparaisons, nous avons défini un coefficient d'affinité qui indique le degré d'attraction ou de répulsion d'une espèce donnée à un brut particulier. Sur les trois crustacés de l'Arctique étudiés, deux amphipodes manifestent une réaction de fuite significative vis-à-vis des masses de pétrole, le type de ce dernier influant de façon marquée sur l'intensité de cette réaction. L'isopode Mesidotea a semblé très indifférent à la présente de brut d'Atkinson Point ou de Vénézuéla. En règle générale, les bruts de l'Arctique se sont révélés moins répulsifs que celui du Vénézuéla.

Selon les premières indications, les adultes d'un certain nombre d'espèces benthiques sont plutôt tolérants à une exposition à court terme à des concentrations modérées de pétrole brut. Etant donné qu'il n'existe pas de populations littorales dans la plupart des régions de l'Arctique du fait du décapage opéré par la glace, le pétrole se déposant sur les plages n'y provoque pas les mortalités massives par étouffement si fréquentes dans les régions tempérées. Parmi les espèces les plus susceptibles d'être gravement endommagées dans les cas d'une

fuite de pétrole, figurent les formes planctoniques et les espèces benthiques qui migrent vers la face interne de la glace en hiver. Le risque potentiel le plus important intervient en effet dans ce cas, lorsque les animaux entrent en contact avec le brut. On a cependant constaté que certaines espèces évitent de façon active ces fuites de pétrole. La réaction de fuite varie à la fois avec l'espèce animale et avec le type de pétrole. Les auteurs passent en revue les autres effets nocifs possibles de la pollution pétrolière sur les populations d'invertébrés marins de l'Arctique, afin d'illustrer l'ampleur du problème qui reste à traiter.

Partie III

a)

Un voyage d'étude de 12 jours a été entrepris dans le sud de la mer de Beaufort, fin juillet 1973, à bord du "North Star of Herschel Island". Il a été impossible d'atteindre plus de la moitié des postes d'échantillonnage, étant donné que la mer était recouverte de glaces à 25 milles du rivage le long de la plupart des côtes. Par conséquent, seules les eaux littorales ont été échantillonnées.

La répartition de la température et de la salinité dans le sud de la mer de Beaufort s'est révélée typique des conditions caractérisant les eaux du large des grands estuaires. Une masse d'eau relativement tiède, de faible salinité, s'étend à partir de l'embouchure du fleuve en dessus d'une couche d'eau plus froide et plus salée. Les composés phosphate-phosphore, nitrate-azote et silicate-silice, qui se sont révélés les plus abondants dans les eaux de surface voisines de l'embouchure, sont manifestement amenés à la mer de Beaufort par le fleuve Mackenzie. Les données relatives à l'oxygène et à la chlorophylle indiquent un rythme de production primaire littorale faible. On peut en conclure que le manque de lumière nécessaire à la photosynthèse, dû à la turbidité importante provenant des eaux du fleuve, constitue probablement le facteur limitant le plus important de la production dans les eaux littorales riches en nutriments.

Au moins 45 espèces de zooplanctons ont été trouvées dans le sud de la mer de Beaufort. Leur nombre pourrait faire croire à une diversité faunique plus grande qu'elle ne l'est en réalité dans la majeure partie de la région. On n'en a trouvé en effet que 13 espèces à 75% des postes, tandis que les deux postes qui comptaient la plus grande diversité en avaient 36. Il semble également que les

stations à faible diversité, relativement nombreuses, comportaient une biomasse réduite. Elles se situaient dans la région où l'influence du Mackenzie s'est révélée maximale au cours de la dernière semaine de juillet 1973, les quelques espèces trouvées (les crustacés Cyclops, Limnocalanus, Eurytemora, Mysis et quelques autres) étant caractéristiques des eaux douces modérémment saumâtres. Quelques-unes de ces espèces provenaient probablement directement du fleuve, survivant au moins pendant un certain temps dans les eaux de surface à faible salinité. La présence en fin de juillet, plus près de l'embouchure, de copepodes à différents stades évolutifs laisse à penser que d'autres espèces peuvent ne pas s'être développées dans les eaux où elles ont été recueillies. Selon plusieurs indications, le taux de production de zooplanctons immédiatement au large de l'embouchure du fleuve est très faible. Plus au large encore, la diversité des espèces s'est révélée plus grande, là où les eaux de l'estuaire sont associées à des eaux "océaniques" et où l'influence du fleuve est probablement insignifiante. La jeunesse des stades évolutifs des crustacés et l'importance apparemment plus grande de la biomasse indiquent un taux de production de zooplancton plus élevé que celui qui peut intervenir plus près des eaux douces.

L'équilibre entre nutriments, plancton végétal et plancton animal est précaire dans la plus grande partie du sud de la mer de Beaufort et dépend du coefficient de mélange des eaux du fleuve ainsi que des influences de celles du large. L'influence du fleuve est à la fois importante et variable. Il ne semble pas que des modifications dépassant les variations annuelles actuelles du débit et des teneurs du fleuve puissent modifier quantitativement et qualitativement sur une surface importante la flore et la faune planctoniques du sud de la mer de Beaufort, immédiatement au large du delta du Mackenzie.

Partie III

b)

Des échantillonnages microbiologiques ont été recueillis au cours d'une tournée d'étude effectuée dans le sud de la mer de Beaufort en 1973. Un comptage du total des organismes viables dans des milieux de culture à base d'eau de mer et d'eau douce a été réalisé, afin d'obtenir une estimation de la répartition et de l'abondance des bactéries hétérotrophiques dans les eaux échantillonnées. On a constaté que le Mackenzie déversait dans la mer de

Beaufort une biomasse importante de bactéries d'eau douce, mais que cette biomasse se dispersait dans les eaux marines. L'abondance des bactéries marines trouvées dans les régions échantillonnées était relativement uniforme. Selon une étude provisoire, cette flore est semblable à celle des lacs des Esquimaux.

CONTENTS

Part I

Degradation of Petroleum by a Psychrotolerant Marine Flora from the Eskimo Lakes, N.W.T.

by

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TABLE OF CONTENTS

LIST OF FIGURES

SUMMARY

In an initial study of biodegradation of petroleum in Arctic marine waters, efforts were made to quantitate and isolate oil-degrading or oleoclastic bacteria in the Eskimo Lakes, N.W.T. The abundance of this flora in the Eskimo Lakes could not be ascertained with the procedure employed. Marine oleoclastic bacteria were, however, isolated by means of an enrichment procedure. In laboratory studies, mixed cultures obtained with the enrichment procedure were found to degrade three petroleum crudes to some extent. Norman Wells crude was degraded at temperatures as low as 2.0°C. Lower temperatures were not evaluated. A preliminary experiment suggested that phosphorus and nitrogen concentrations in seawater might not be limiting for degradation at low temperatures, but further studies are required.

INTRODUCTION

> "Of all of the scientific and technological problems associated with sea pollution the most important is the capacity of the oceans for the biochemical degradation of wastes. Until this is better understood, pollution abatement planning cannot be scientifically undertaken, nor in the judgment of the author, will sea pollution by oil be brought under control." J.E. Moss (1971)

The ability of microorganisms to degrade hydrocarbons and fractions of crude petroleum has been well documented over the last thirty years. The first comprehensive review of the subject was prepared by ZoBell in 1946. Much information has been gained in studies of biodegradation of petroleum in the marine environment, and recent reports have speculated on the possibility of similar biodegrading processes in the Arctic ecosystem.

This interest has resulted from the ever-expanding exploration for oil and gas in the Canadian and American Arctic. Off-shore drilling as well as the possible translocation of discovered oil by tanker or pipeline present the probability of spillage in the Arctic marine environment.

Difficulties encountered with techniques, facilities and transportation have resulted in the neglect of the microbiology of Arctic marine waters in the past. This study was possible because of the unique facilities and logistical resources of the Arctic Biological Station in the Western Canadian Arctic. The study was initiated to assess the possibility of petroleum biodegradation in Arctic marine waters by an indigenous microbial flora as a part of a long-term study of Arctic marine ecology presently being conducted by this station.

This initial report deals with the results of work begun in July, 1973, within the framework of the following objectives:

a) quantitation of the bacterial populations in

marine waters which were capable of degrading petroleum;

b) isolation and characterization of oil-degrading or oleoclastic bacteria;

c) determination of the capability of mixed and pure cultures of oleoclastic cells to degrade various crude petroleums at different temperatures;

d) determination of limiting factors such as nitrogen and phosphorus in the biodegradation of petroleum.

METHODS AND SOURCES OF DATA

1. Total Viable Counts of Heterotrophs

(a) Eskimo Lakes

Water samples from various depths at Station 508 (see Figure 1) were collected aseptically with Niskin SS 1.5 sterile bag-samplers from a freighter canoe or float-plane prior to disturbances of the water column by other oceanographic samplings. Water samples collected in this manner were aseptically transferred to cold, sterile polypropylene bottles (one litre) and flown to laboratory facilities in Inuvik, N.W.T., in a chilled condition, where they were processed within four hours of collection from the water column. To enhance the multiplication and colony formation of marine bacteria, ZoBell Marine Broth 2216E (Difco), containing a formulation of seawater salts, was dissolved in deionized water and solidified with 1.2% Bacto-agar (Difco). A spin-plate technique was employed to dispense an aliquot of water sample with a cold pipette on the surface of a cold agar plate which was then placed in a 5.0°C incubator. Upon absorption of the aliquot of water by the agar medium, the plate was inverted and incubation was continued at 5.0°C for three weeks. Quadruplicate spin-plates were made of each water sample. After incubation, the plates were examined and those with an uneven distribution of colonies were discarded. The colonies of three plates of a replicate

set were enumerated, averaged, and the mean value was expressed as the log number of colony-forming units (C.F.U.) per one litre of water sample.

(b) South Beaufort Sea

Microbiological samplings were conducted during a cruise on the South Beaufort Sea in July, 1973 (see Part Three in this series of reports). The procedures described in the previous section were employed to collect and process samples aboard the vessel, "North Star of Herschel Island".

2. Membrane Filter Enumeration of Oleoclastic Heterotrophs

At the same time ZoBell spin-plates were being inoculated, 50.0 and 100.0 ml aliquots of Eskimo Lakes water samples were filtered through sterile 0.45 μ (47 mm dia.) membrane filters contained in sterile, cold Sterifills (Millipore Corp.). Replicate filters were placed on the cold agar surface of carbon-free artificial seawater plate media (Instant Ocean) with and without 1.0% weathered, emulsified Norman Wells crude. Filters placed on ZoBell plate media served as controls. Plates were inverted and incubated at 5.0°C until colonies developed on the filters.

3. Oil Enrichment Broths

Five hundred ml aliquots of Eskimo Lakes water samples were filtered in the manner described above and the filters were transferred to acid-washed, sterile, 500 ml Erlenmeyer flasks containing 100.0 ml of Eskimo Lakes water supplemented with 1.0 gm NH_4NO_3, 0.5 gm K_2HPO_4, 7.69 gm Trizma (Sigma Chemical Co.) and 10.0 ml weathered Norman Wells crude per litre. Final pH at 5.0°C was 7.80. Cultures were agitated on a gyratory shaker at 200.0 rpm in a 5.0°C incubator until visible emulsification occurred after about four weeks. Cultures were then transferred in a chilled condition to the Arctic Biological Station, Ste. Anne de Bellevue, Québec.

4. Petroleum Degradation Experiments

(a) Media

For viable count determinations, ZoBell marine agar was employed (see before). An oil-seawater broth was employed for the maintenance of cultures and all experiments. Half-strength artificial seawater (salinity ca. 17‰) was prepared by dissolving a commercial sea salt mixture (Instant Ocean) in distilled water. The half-strength seawater was supplement with 1.0 gm NH_4NO_3, 0.5 gm K_2HPO_4 and 7.69 gms Trizma per litre unless otherwise indicated, dispensed in 400 ml aliquots in 1.0 l Erlenmeyer flasks and autoclaved. After cooling, the medium was supplemented with 0.4 ml of a filter-sterilized, weathered crude oil. Final pH at 5.0°C was 7.80. Crude petroleums used in these experiments were weathered under a forced air draft for ten days in 150 mm glass petri dishes. After weathering, residual crudes were filter-sterilized with 0.45 μ membrane filters (Millipore) and stored at 5.0°C in glass vials with teflon-lined caps until required.

N.B. All glassware used in these experiments was acid-washed with chromic acid and carefully rinsed with distilled water.

(b) Inocula

One of the enriched oil cultures obtained from the Eskimo Lakes was used in all experiments. The mixed culture was maintained by subculturing into fresh oil-seawater broth, and incubating at 5.0°C on a gyratory shaker at 200 rpm. Four ml of a six-day culture were employed as an inoculum for experiments.

(c) Viable Counts

In an experiment, viable counts of all cultures were determined. A one ml aliquot of a culture was serially diluted to 10^{-7} in ten-fold dilutions. A spin-plate technique was employed to inoculate 0.1 ml of each dilution on triplicate plates of ZoBell Marine Agar. Plates were incubated at 15°C for five days and the plates prepared from one dilution were chosen for enumeration. Plates of a replicate set were enumerated, averaged, and the mean value was expressed

as the log number of colony-forming units (C.F.U.) per one ml of culture.

(d) Extraction of Residual Petroleum from Cultures

See Appendix, No. 1.

(e) Gas Chromatographic Analysis of Extracted Petroleum

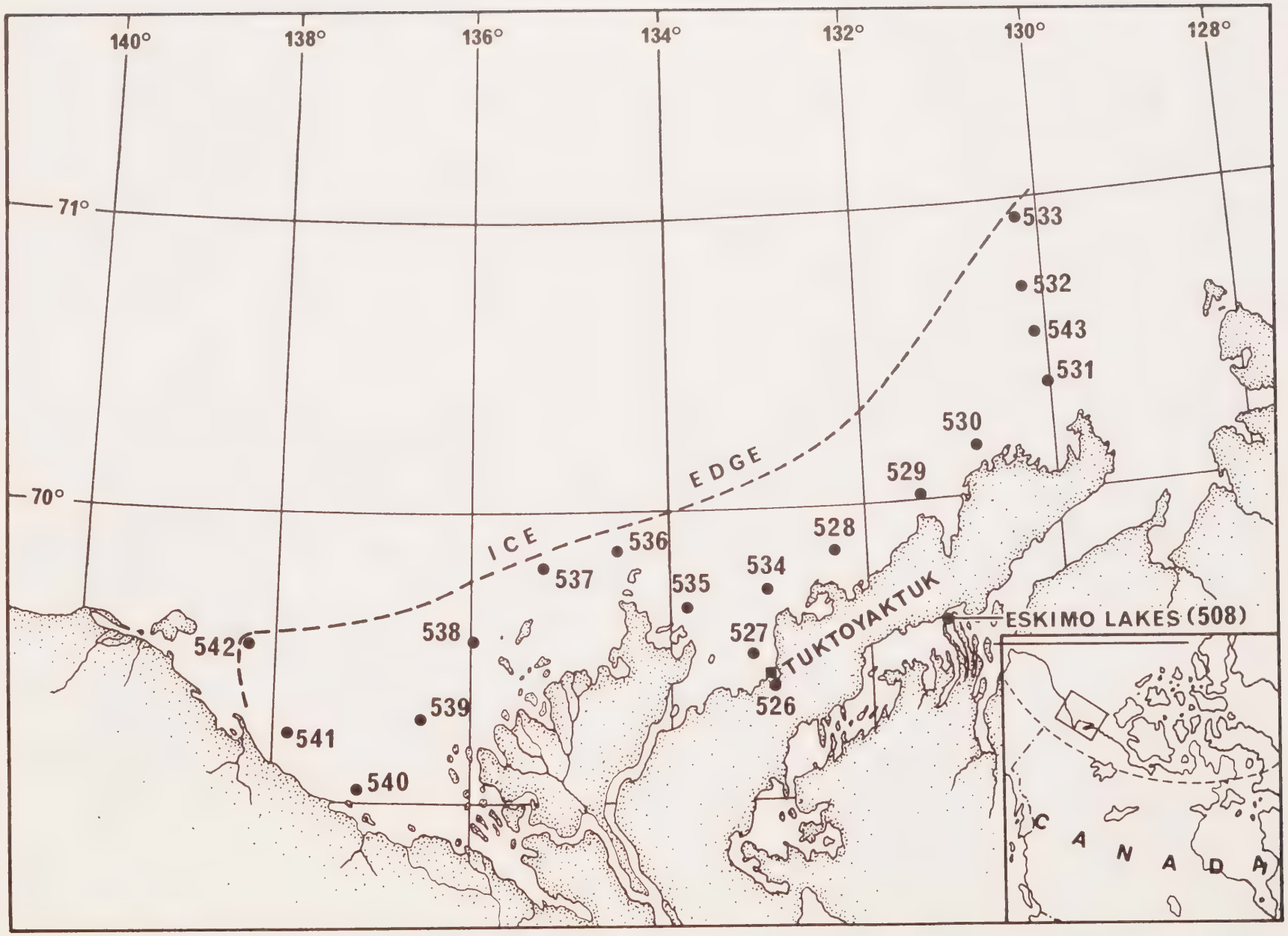
See Appendix, No. 2.

RESULTS

1. Total Viable Counts in the Sampled Areas

In a recent cruise on the South Beaufort Sea during July, 1973 (see report 3 in this series), the total viable counts of marine heterotrophic bacteria were determined at stations occupied (Figure 1). The abundance of heterotrophs was found to be similar to or in excess of counts obtained at a more intensively sampled station (508) in the Eskimo Lakes during the same period. Values of 10^6 to 10^7 counts per litre of waters in these areas compare favourably with values reported by investigations of estuaries and offshore areas along the New England coast (Kaneko and Colwell, 1973; Sieburth, 1967; Atlas and Bartha, 1972a). In drawing such comparisons, however, varying methods of sampling and cultivation must be considered.

In the Eskimo Lakes, an inlet of the Beaufort Sea, seasonal samplings on open water and through the winter ice have demonstrated that the total viable count of marine heterotrophs varies by about two orders of magnitude across the year (Figure 2). Counts obtained by incubation of plates at 5.0° and 15.0°C suggest that a large population of obligately psychrophilic heterotrophs is present. Characterization of representative isolates taken from cultured plates at various times of the year reveal that the majority of isolates from plates incubated at 5°C fail to grow above 20.0°C and have a temperature optimum for growth and multiplication at 10.0 to 15.0°C. These heterotrophs are considered to be obligately psychrophilic. Isolates from plates incubated at 15.0°C have been characterized as obligate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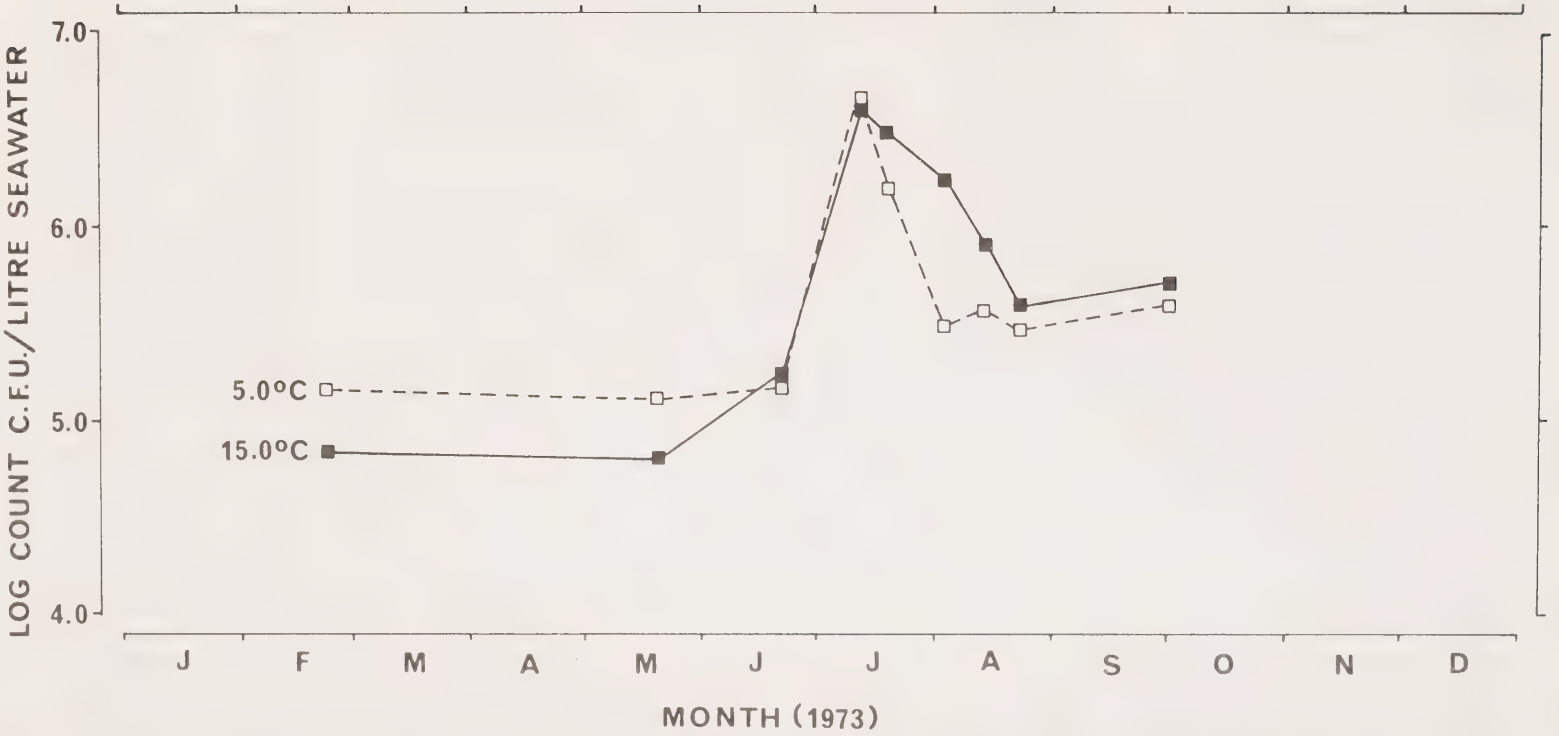

Figure 2. Seasonal distribution of marine heterotrophs at 10 metre level of Station 508 in the Eskimo Lakes. Inoculated plates of ZoBell marine agar were incubated at 5.0° and 15.0°C. Plates of a replicate set were enumerated, averaged and the mean value was expressed as the log number of colony-forming units (C.F.U.) per one litre of water sample. Log C.F.U. values are plotted against the date of sampling. Station 508, located at the neck of the Eskimo Lakes, is characterized by strong tidal currents and demonstrates a relatively homogenous water column of twenty metres.

Salinity range - ca. 12 to 17°/$_{oo}$
Temperature range - ca. -1.2 to 9.0°C
Ice cover - late November to late June up to 170 cm.

psychrophiles as well as psychrotolerant cells which fail to multiply above 30.0 to 35.0°C.

2. Estimation of Oleoclastic Heterotrophs in the Eskimo Lakes

Attempts to enumerate an oleoclastic population in the Eskimo Lakes were without success. Large numbers of colonies grew on the membrane filters in the presence or absence of oil in the carbon-free medium. Although membrane filtration of seawater concentrated heterotrophs on the filter, it also resulted in the concentration of micro-detritus. Heterotrophs unable to use the petroleum supplemented in the medium as a sole carbon source for multiplication were probably able to utilize the concentrated detritus as a carbon source. The abundance of oleoclastic heterotrophs in the Eskimo Lakes is considered to be too low to make use of a "most probable number" technique. At this moment, a method of enumeration for these organisms remains to be devised.

3. Oil Enrichment Cultures

Visible emulsification was observed in the enrichment cultures after four weeks of incubation at 5.0°C with continuous agitation. It is important to remember that the inoculum for the 100 ml oil enrichment broths consisted of bacteria concentrated from 500 ml of Eskimo Lakes water from Station 508 on a membrane filter. The long lag before visible emulsification was indicative of the paucity of oleoclastic heterotrophs in the 500 ml seawater sample. One enriched culture was subcultured three times at monthly intervals into a fresh oil-seawater broth and in December, 1973, the mixed culture of oleoclastic bacteria obtained by the enrichment procedure was employed as the inoculum for subsequent experiments described in this report.

4. Degradation of Norman Wells Crude by a Mixed Culture at Defined Temperatures

It had been anticipated that obligately psychrophilic organisms would be found in the mixed culture because of the low temperature isolation procedure. Preliminary data, however, indicate that the eight

bacterial types in this culture are all psychrotolerant.

To demonstrate the effect of temperature on the degradation of Norman Wells crude by the mixed culture, identical oil-seawater broths with and without inocula were incubated at 2, 5, 10 and 15°C. Replicate cultures and oil controls at the four temperatures were removed at intervals and their residual oils were extracted. Relative losses in the aliphatic fraction were calculated by measuring the area under the GC profile peaks of the 14C, 15C, 16C, 19C, 20C, 21C and 22C saturated hydrocarbons with a planimeter. Peaks were identified by comparison with known standards. These well-defined peaks were considered to be representative of the total profile of the aliphatic fraction after weathering and errors introduced by measuring poorly-defined peaks were avoided. The values of the summed peak areas were expressed as a percentage of the values obtained from the profiles of oil-control vessels incubated in an identical fashion but with inocula deleted. The results are seen in Figure 3. A continuous increase in the rate of degradation was observed with increasing temperature up to and including 20.0°C (data not reported). At 2.0°C, a noticeable lag occurred before degradation commenced, but no appreciable lag was observed at the higher temperatures. Four of the GC profiles employed in the construction of the 15°C curve (Figure 3) are seen in Figure 4.

5. Response of a Mixed Culture to Several Crudes

Petroleum crude is a complex mixture of many hydrocarbons and crudes from different geographical regions vary in composition. For this reason, the response of an oil-degrading culture to different crudes may vary, depending on the types and concentrations of hydrocarbon substrates available for oxidation. For this experiment, two indigenous northern crudes, from Norman Wells, N.W.T. and Atkinson Point on the Tuktoyaktuk Peninsula, N.W.T., were obtained from Imperial Oil Ltd. (Calgary). Pembina crude from Northern Alberta was obtained through the Freshwater Institute (Winnipeg). GC profiles of these oils are shown in Figure 5.

Analysis of residual petroleum after incubation of the three oils at 15°C with the mixed culture for various periods demonstrated a reduction in the aliphatic profiles of the Norman Wells and Pembina crudes (Figure 6).

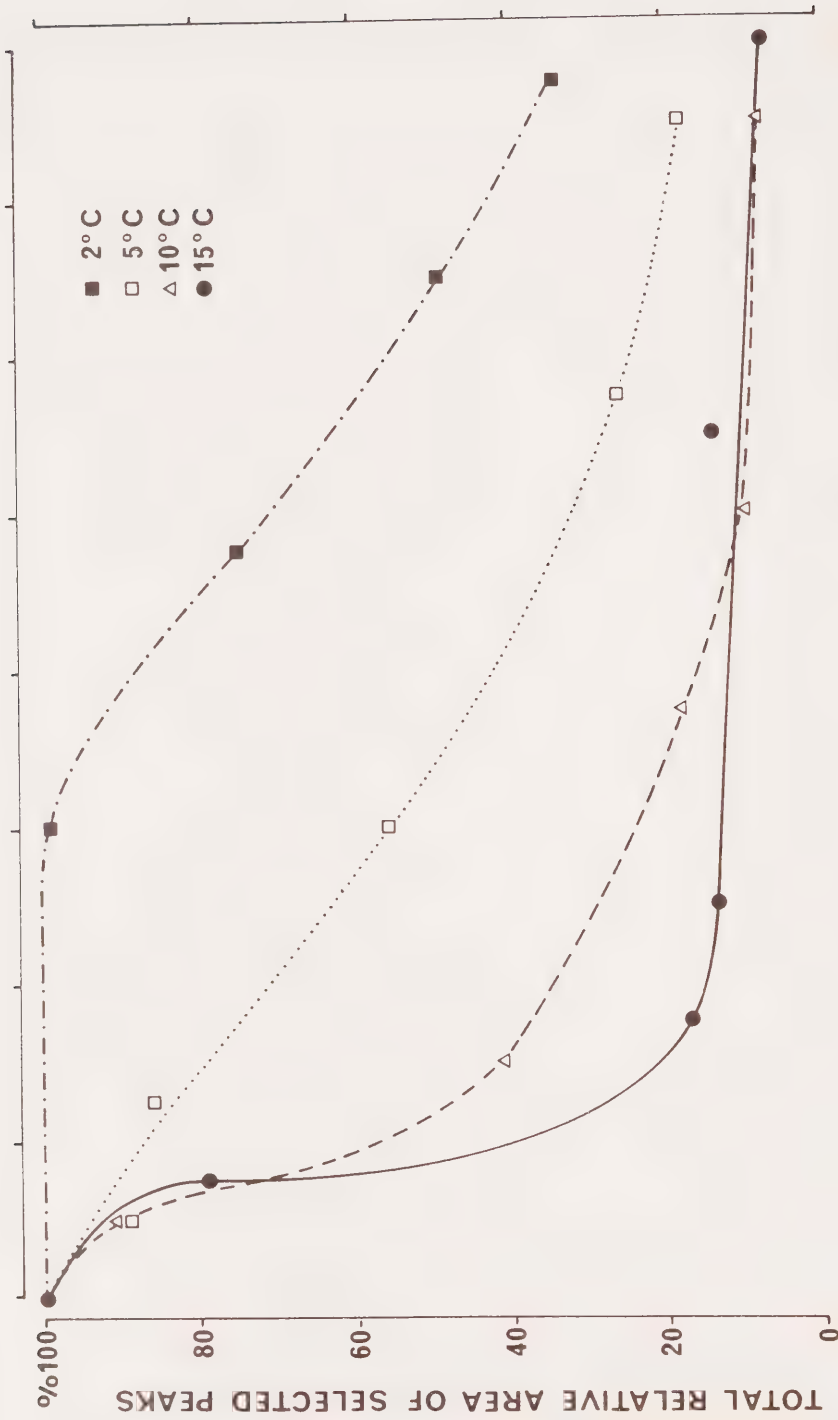

Figure 3. Degradation of weathered Norman Wells crude by a mixed culture at defined temperatures. Cultures and oil controls were removed at intervals during incubation and residual petroleum was extracted from each. Summed areas of selected peaks of GC profiles were expressed as percentages of the summed areas of the same peaks in undegraded controls. Percentage values obtained are plotted against incubation interval when extracted.

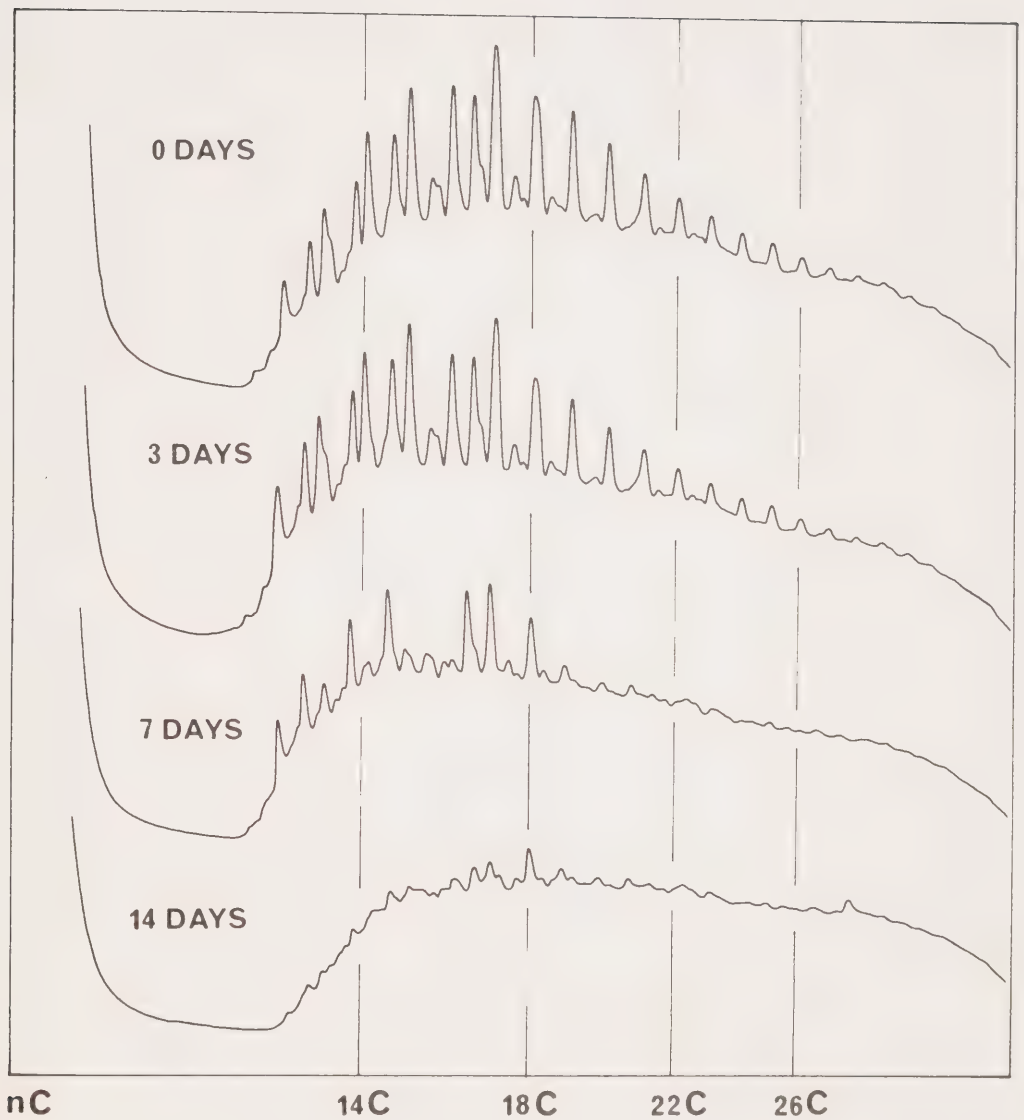

Figure 4. GC Profiles of residual petroleum extracted from four cultures incubated at 15°C for various intervals. Carbon numbers (nC) indicate peaks of several saturated hydrocarbons. Values calculated from these profiles were employed in the construction of the 15.0°C curve in Figure 3.

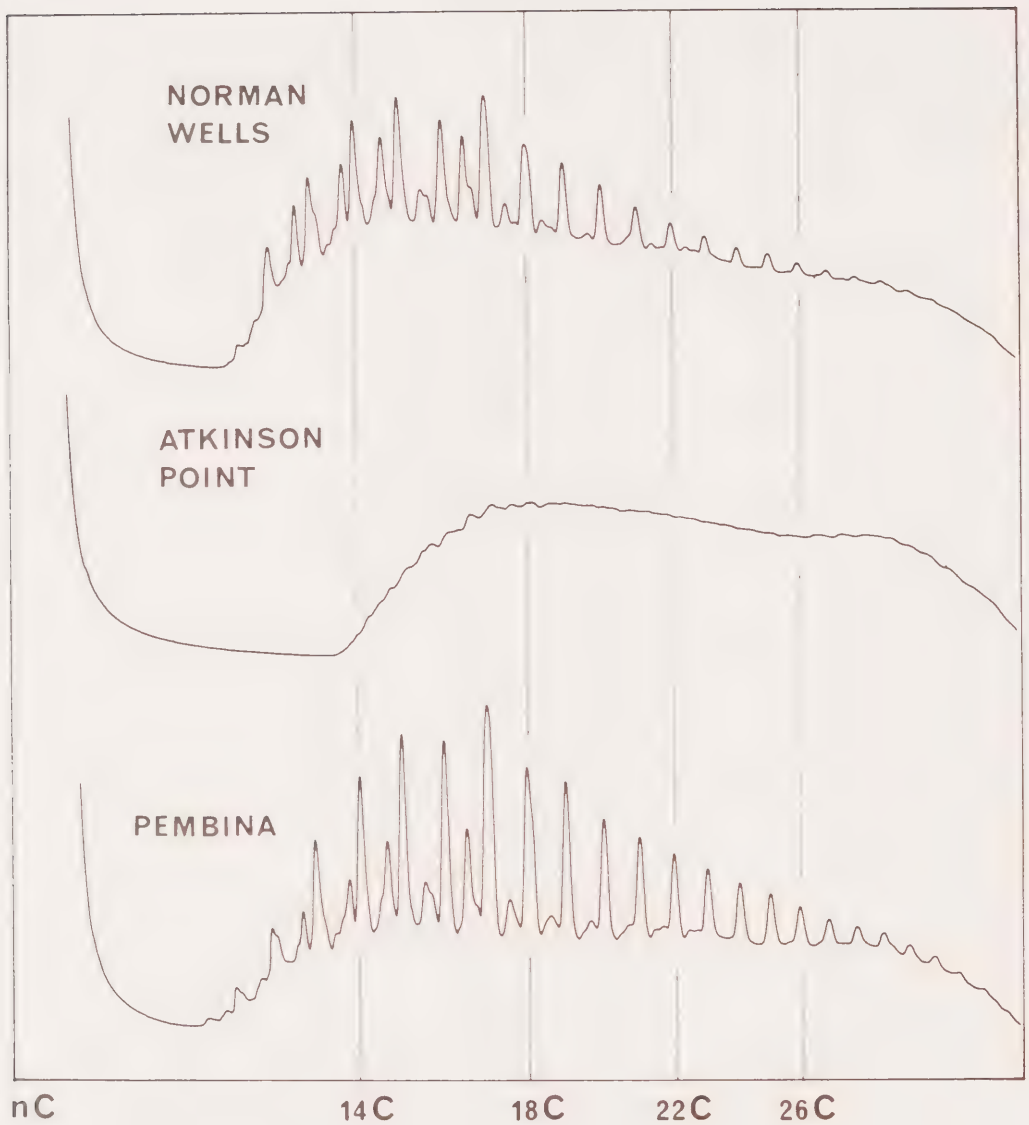

Figure 5. GC Profiles of the three weathered crudes used in these experiments. Carbon numbers (nC) indicate peaks of several saturated hydrocarbons.

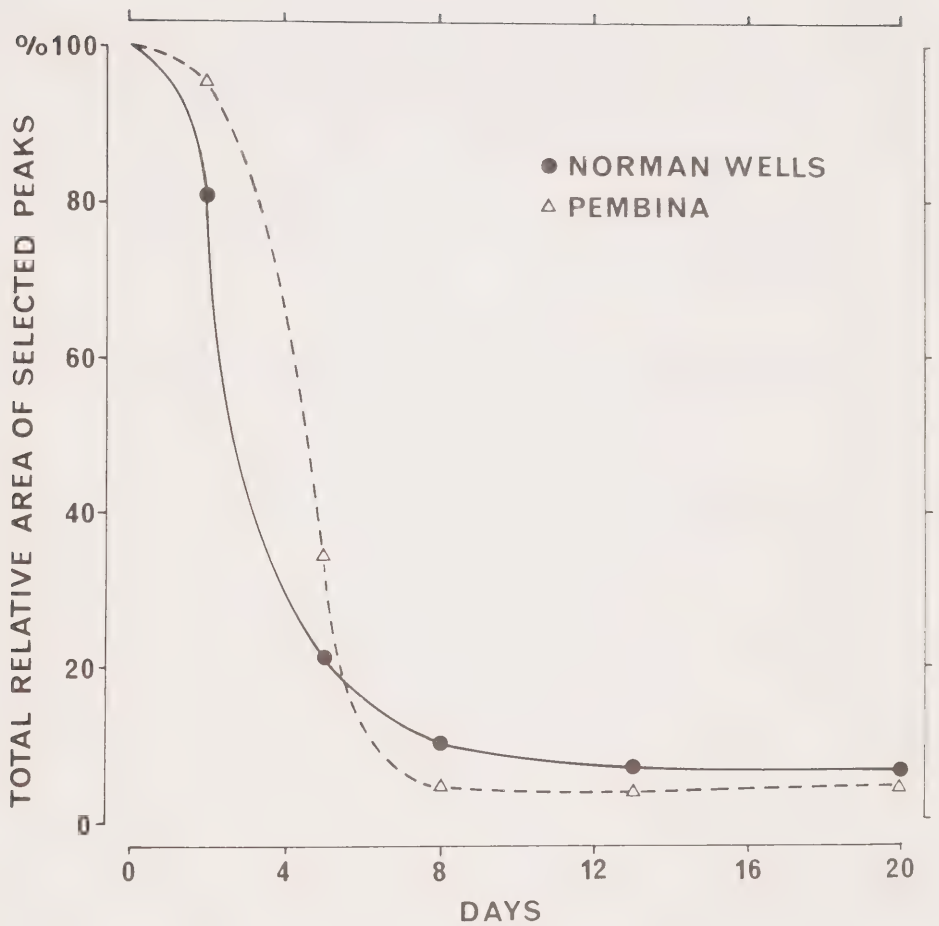

Figure 6. Degradation of weathered Norman Wells and Pembina crudes by a mixed culture at 15°C. See legend to Figure 3.

The degradation of Atkinson Point crude, a petroleum low in aliphatic content, could not be evaluated with our procedure. A degree of mineralization of all three oils, however, was indicated by a preliminary gravimetric procedure. Viable counts taken before extraction of each culture demonstrated an increase in biomass parallel to the rate of loss of the aliphatic fractions of Norman Wells and Pembina crudes (Figure 7). A low concentration of aliphatic substrate in the Atkinson Point crude may have resulted in the lower biomass of this oil culture compared to those observed with the other oil cultures at the end of the experiment. The increase in count of the Atkinson Point culture, however, suggested a degree of utilization of this oil. In the absence of a carbon source, the oil-free control culture did not show an increase in count.

6. Effect of Deletion of Nutrient Supplements from the Oil-Seawater Medium on Degradation by a Mixed Culture

In the isolation and cultivation of the mixed culture, NH_4NO_3 and K_2HPO_4 were routinely supplemented in the oil-artificial seawater medium. To determine the effectiveness of these supplements, degradation of Norman Wells crude at 15°C in cultures supplemented with NH_4NO_3 or K_2HPO_4 or both were compared to degradation in unsupplemented cultures. Cultures and oil controls were removed at intervals and the residual petroleum of each was extracted and assayed. The results were seen in Figure 8. A similar degree of degradation was found in the presence or absence of NH_4NO_3 and K_2HPO_4 but, inexplicably, less degradation of the aliphatic fraction was observed where K_2HPO_4 was present and NH_4NO_3 absent. The increase in viable count in all cultures paralleled the loss of the aliphatic fraction (Figure 9). The results suggest that phosphorus and nitrogen may not be limiting in the artificial seawater medium and supplements may not be required in order for degradation of Norman Wells crude by the mixed culture to proceed at 15°C. However, phosphorus and nitrogen requirements for degradation of petroleum by pure cultures selected from the mixed culture remain to be determined in a simplified, defined seawater medium.

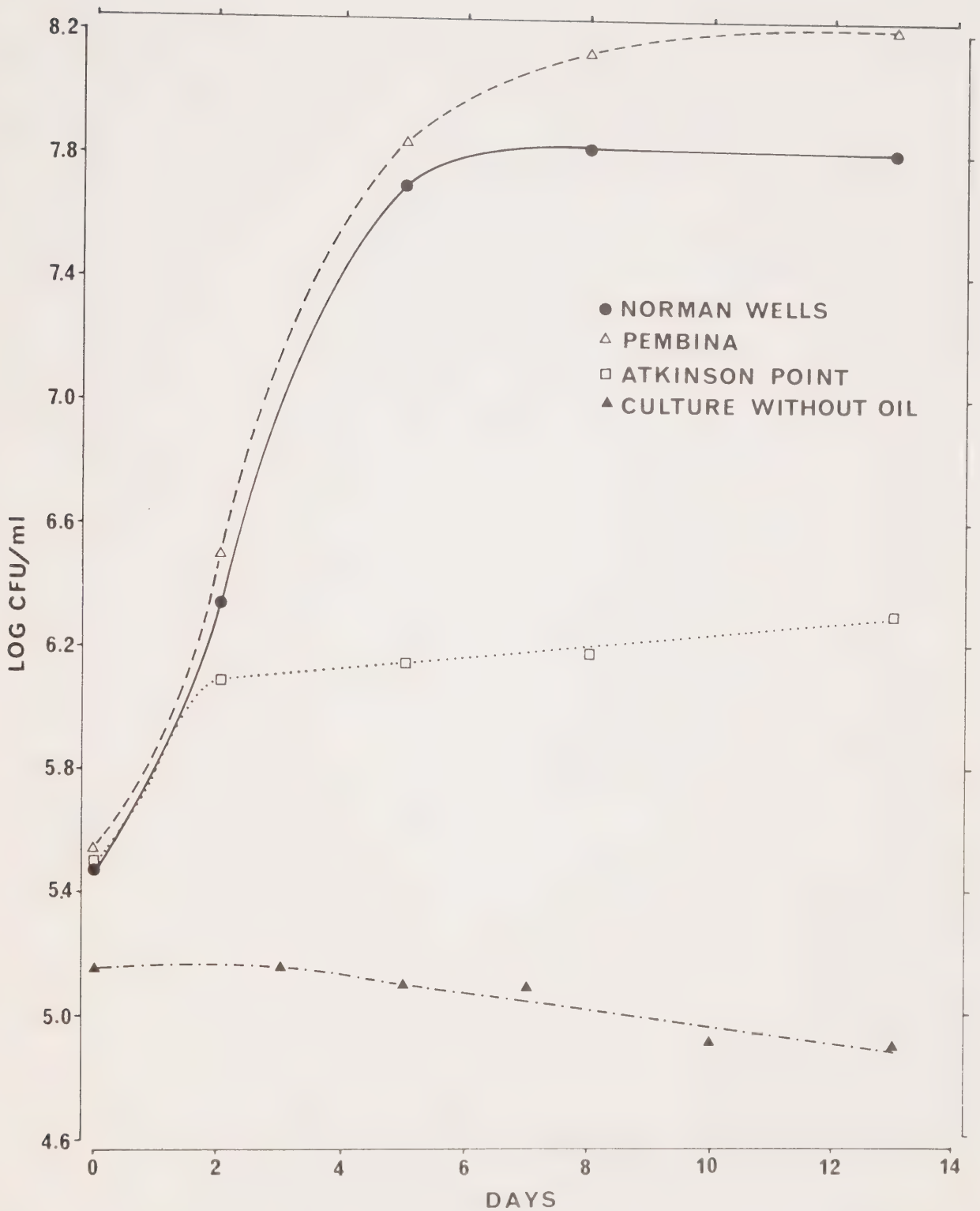

Figure 7. Increase in viable counts of oil and control cultures during degradation of weathered Norman Wells and Pembina crudes by a mixed culture at 15°C. Values of extracted petroleum from these cultures are plotted in Figure 6. Viable counts are expressed as log colony-forming units (C.F.U.) per 1.0 ml of culture and are plotted against incubation interval when sampled.

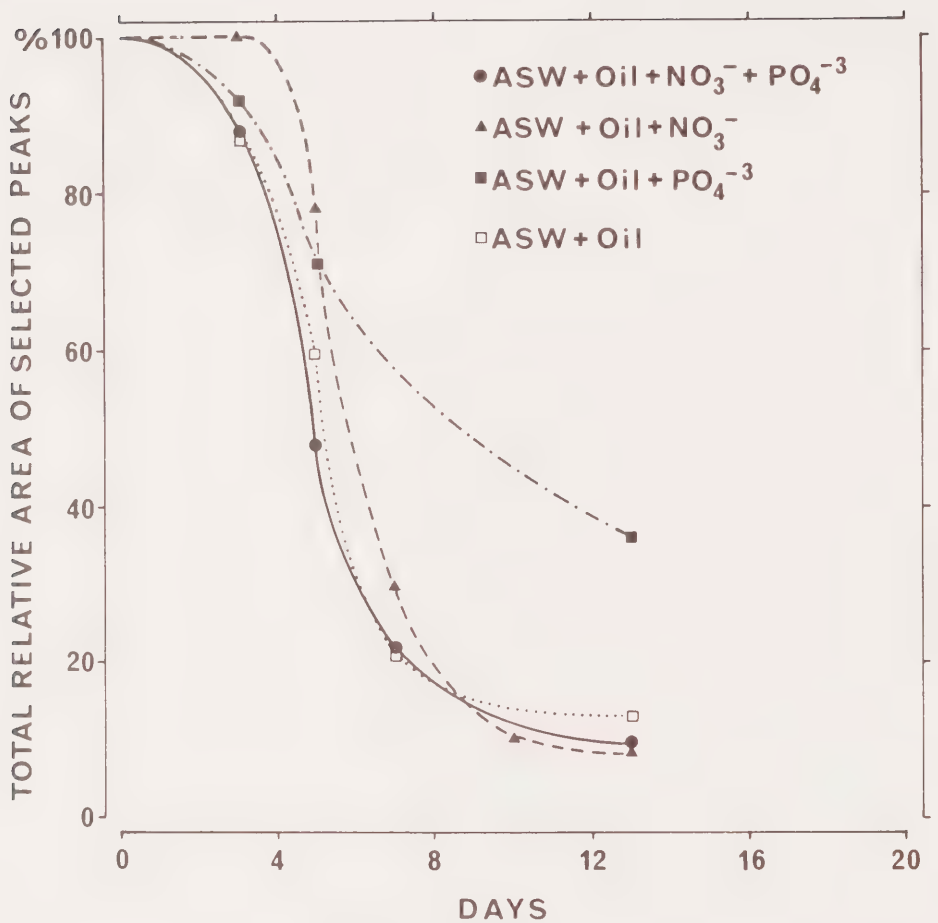

Figure 8. Degradation of weathered Norman Wells crude by a mixed culture at 15°C in the presence or absence of phosphorus and nitrogen supplements. See legend to Figure 3. ASW refers to the artificial seawater medium.

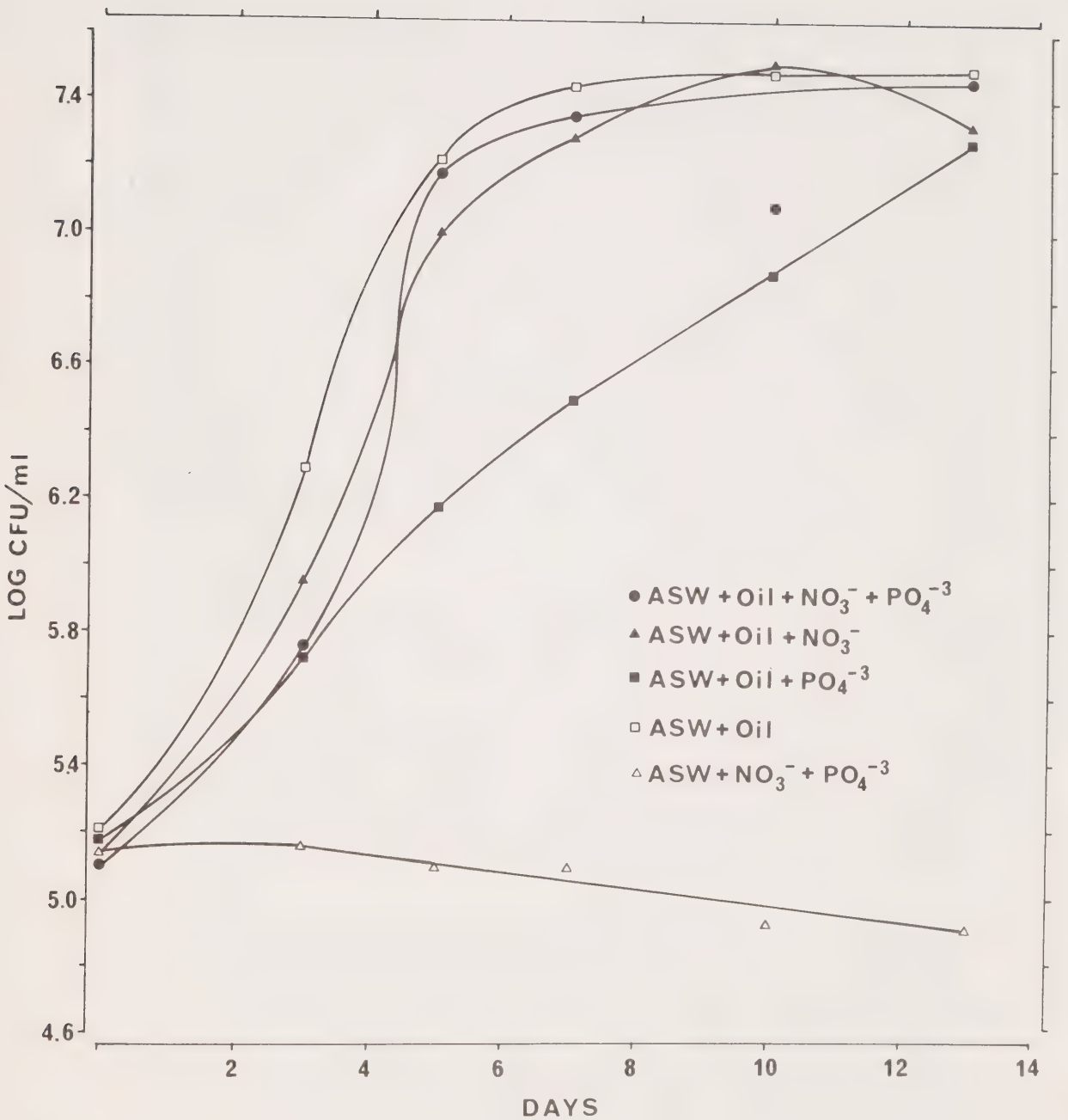

Figure 9. Increase in viable counts of oil and control cultures during degradation of weathered Norman Wells crude by a mixed culture at 15°C in the presence or absence of phosphorus and nitrogen supplements. Values of extracted petroleum from these cultures are plotted in Figure 8. Viable counts are expressed as log colony-forming units (C.F.U.) per 1.0 ml of culture and are plotted against incubation interval when sampled. ASW refers to the artificial seawater medium.

DISCUSSION

In recent years, much attention has been given to the fate of petroleum and petroleum products in various ecosystems. Biodegradation of these substances by indigenous microorganisms has been demonstrated in many cases. Petroleum biodegradation in the marine environment has been shown by a number of laboratory and field studies and these investigations have emphasized the importance of such environmental parameters as temperature, oxygen concentration and nutrient supply in determining the rate of biodegradation. After experiments with winter samples of the indigenous marine flora of the New Jersey coastline, Atlas and Bartha (1972a) have suggested that the biodegradation of petroleum in marine waters will be appreciably reduced at low temperatures. In an Arctic marine ecosystem, however, where low temperatures are prevalent throughout the year, the response of an indigenous flora of bacterial heterotrophs to the presence of petroleum would be dependent on the abundance and diversity of this flora and above all, its metabolic activity at low temperatures.

Although attempts to enumerate the oil-degrading or oleoclastic heterotrophs in the Eskimo Lakes last summer were unsuccessful, their isolation was accomplished by an enrichment procedure. The marine bacteria isolated in this manner degraded petroleum at temperatures as low as 2.0°C, but optimum growth and multiplication of the individual components of the mixed culture occurred at 20.0 to 25.0°C in an organic medium (data not presented) and indicated that the isolates were psychrotolerant rather than obligately psychrophilic.

The initial experiments reported here were designed to demonstrate the biodegradation of several crudes at various temperatures by the mixed psychrotolerant culture. The measurement of the relative loss of the aliphatic fraction was employed as the main criterion of microbial modification of the crudes. Berridge et al. (1968) have summarized the chemical and physical characteristics of various crudes and emphasized their dissimilarities. The compositional differences of the three crudes employed in the present study were demonstrated by preliminary gravimetric determinations and the response of the mixed culture

to each crude. Similar viable counts were obtained during the degradation of Norman Wells and Pembina crudes and similar rates of loss of the aliphatic fractions were observed. Residual weights of the oils after degradation, however, suggested that the Pembina crude was less susceptible to microbial degradation. On the other hand, Atkinson Point oil, low in aliphatic content, supported multiplication of the mixed culture to some extent, but degradation could not be assessed by gas chromatography. These initial results indicate that in addition to the present procedures, gravimetric determinations are required to quantitate the mineralization of various petroleums in the culture vessels.

Various reports in the literature have noted that oil-enriched cultures must be supplemented with nitrogen and phosphorus. Bartha and Atlas (1972b) found nitrogen and phosphorus in seawater to be severely limiting to the process of oil degradation. On the other hand, Kinney et al. (1969) reported that the concentrations of these nutrients in seawater were not limiting at lower temperatures due to the reduced requirements of a slower rate of biodegradation. When the nutrient supplements were deleted from the artificial seawater used in the present study, the rate of biodegradation did not appear to be affected in one instance, but the results require further substantiation. Further studies are required with a simplified, defined seawater medium to assess the relevance of nutrient supplements.

The rapid degradation of the aliphatic fractions of Norman Wells and Pembina crudes by the mixed culture does not reflect in situ activity. In natural waters, the lag before biodegradation would be determined by the abundance of oleoclastic heterotrophs and their degree of activity at low temperatures.

Robertson et al. (1973) reported that plate enumerations did not reveal any oleoclastic heterotrophs in estuarine areas of the Colville River near Point Barrow, Alaska. However, ZoBell (1972) has determined that obligately psychrophilic floras in oil-soaked tundra muck and oil-contaminated waters from the same region of Alaska degraded petroleum. Samplings of the obligately psychrophilic marine population of the Eskimo Lakes are being continued to isolate oleoclastic bacteria. These efforts may be hindered by the possibly low abundance of these cells in an ecosystem which has remained free of petroleum contamination.

CONCLUSIONS

1. A marine flora of heterotrophic bacteria has been established to exist at a relatively uniform level of abundance in the sampled areas of the South Beaufort Sea and the Eskimo lakes.

2. A population of oleoclastic cells exists within the heterotrophic marine flora of the Eskimo Lakes but the level of abundance of these cells may be too low to be assessed quantitatively.

3. Psychrotolerant cells isolated from the Eskimo Lakes by an enrichment procedure are capable of degrading weathered Norman Wells crude at temperatures as low as 2.0°C. Degradation at lower temperatures was not evaluated.

4. Isolated cultures are capable of utilizing Norman Wells, Atkinson Point and Pembina weathered crudes for growth and multiplication. In each instance, microbial modification of the crude was observed.

5. Phosphorus and nitrogen at the concentrations found in seawater may not be limiting for biodegradation to proceed at low temperatures. This conclusion, however, is tentative and subject to the completion of the study.

6. An oleoclastic potential probably exists in the Eskimo Lakes, and by implication, in the South Beaufort Sea, but *in situ* rates of biodegradation of petroleum cannot be estimated at this time.

IMPLICATIONS AND RECOMMENDATIONS

The results of preliminary field and laboratory studies tend to suggest the possibility of biological degradation of petroleum in Arctic marine waters but further studies are required. The persistence of petroleum in the Arctic marine ecosystem cannot be ascertained at this time. However, some microbial modification of weathered, residual petroleum might be expected.

NEEDS FOR FURTHER STUDY

To increase knowledge of biological degradation of petroleum in the Beaufort Sea - Eskimo Lakes ecosystems, further studies are required to:

1. quantitate the abundance of oleoclastic heterotrophs;

2. determine the minimum and optimum temperatures for biological degradation by various heterotrophic isolates;

3. determine concentrations at which phosphorus and nitrogen become limiting for biodegradation under defined conditions;

4. devise procedures for measuring the *in situ* rate of biodegradation in the Arctic marine ecosystem.

REFERENCES

Atlas, R.M., and R. Bartha. 1972a.
Biodegradation of petroleum in seawater at low temperatures. Canad. J. Microbiol. 18: 1851-1855.

Bartha, R. and R.M. Atlas. 1972b.
Biodegradation of oil in seawater; limiting factors and artificial stimulation. In: D.G. Ahearn and S.P. Meyers (eds). The Microbial Degradation of Oil Pollutants. pp. 147-152. Center for Wetland Resources. Publication No. LSU-SG-73-01. Louisiana State U., Baton Rouge, La.

Berridge, S.A., R.A. Dean, R.G. Fallows and A. Fish. 1968.
The properties of persistent oils at sea. J. Inst. Petrol. London. 54: 300-309.

Keneko, T. and R.R. Colwell. 1973.
Ecology of Vibrio parahaemolyticus in Chesapeake Bay. Journ. Bacteriol. 113: 24-32.

Kinney, P.J., D.K. Button, and D.M. Schell. 1969.
Kinetics of dissipation and biodegradation of crude oil in Alaska's Cook Inlet. In: Proc. API/FWPCA Joint Conf. on Prevention and Control of Oil Spills. pp. 333-340 API Publ. No. 4040.

Moss, J.E. 1971.
Petroleum - the problem. In: D.W. Hood (ed). Impingement of Man on the Oceans. pp. 381-419. J. Wiley and Sons. New York.

Robertson, B., S. Arhelger, P.J. Kinney and D.K. Button. 1973.
Hydrocarbon biodegradation in Alaskan waters. In: D.G. Ahearn and S.P. Meyers (eds). The Microbial Degradation of Oil Pollutants, pp. 171-184. Center for Wetland Resources. Publication No. LSU-SG-73-01. Louisiana State U., Baton Rouge, La.

Sieburth, J. McN. 1967.
Seasonal selection of estuarine bacteria by water temperature. J. exp. mar. Biol. Ecol. 1: 98-121.

ZoBell, C.E. 1946.
Action of microorganisms on hydrocarbons. Bact. Rev. 10: 1-49.

Zobell, C.E. 1972.
Bacterial degradation of mineral oils at low temperatures. In: D. G. Ahearn and S. P. Meyers (eds). The Microbial Degradation of Oil Pollutants. pp. 153-161. Center for Wetland Resources. Publication No. LSU-SG-73-01. Louisiana State U. Baton Rouge, La.

APPENDIX

1. Extraction of Residual Petroleum from Cultures

Extraction of residual petroleum from culture vessels was accomplished with three 20 ml aliquots of n-pentane in a 500 ml separatory funnel. The n-pentane-oil partitioned above the aqueous layer was removed each time with a pipette. The combined fractions were added to a tared 100 ml beaker and evaporated to dryness at room temperature. When no further weight loss was noted, the weight of oil was recorded and a 40.0 µg/µl solution of the oil was prepared with n-hexane. This solution was stored in teflon-capped glass vials until analysed.

2. Gas Chromatographic Analysis of Extracted Petroleum

A Hewlett-Packard model 5711A gas chromatograph was used for analysis of the oil samples. A 5.0 µl sample of the extract-hexane solution was injected into a 10.0 foot stainless steel column (0.125 in O.D.) packed with 3.0% OV-1 on 100-120 mesh Chromasorb P (Chromatographic Specialties Ltd.). Dual flame ionization detectors were supplied with an air-flow rate of 240 ml/min at 24 psi and a hydrogen-flow rate of 30 ml/min at 14 psi. Nitrogen carrier gas was regulated to 20 ml/min at 60 psi. Oven temperature was programmed for an initial temperature of 70.0°C and increased at a rate of 8.0°/min to 320.0°C where it was held for eight minutes. The readout, attenuated X160, was made on a single-pen Fisher recorder with chart-speed adjusted to 0.5 in/min. Peak areas were calculated with a planimeter.

Part II

Effects of Crude Oil on Arctic Marine Invertebrates

by

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February 1974

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

SUMMARY

Little is known about the short- or long-term biological consequences of oil pollution upon Arctic marine communities. On the basis of considerable research carried out in temperate regions it has been predicted that effects upon Arctic ecosystems would be particularly severe, especially from the point of view of rate of recovery from damage. A survey of the extensive literature dealing with the impact of oil upon marine invertebrates in general, is presented in order to indicate the potential magnitude and nature of the petroleum hazard to marine life and to set the present study in context.

This study is designed to yield information about potentially harmful interactions between some of the dominant marine invertebrates found in shallow coastal waters in the vicinity of the MacKenzie Delta, and Arctic crude oil. Two Arctic crude oils have been utilized, one from Norman Wells and the other from the Atkinson Point Well, located on the Tuktoyaktuk Peninsula adjacent to the study area. A sample of Venezuelan crude (an oil that has received considerable study regarding its biological impact) has recently been obtained and used for comparative purposes in some experiments.

Crude oil may directly damage organisms in two distinct ways. Certain of its components may damage tissues by chemically interfering with cell structure or function. In addition, the whole oil itself, by virtue of its high viscosity, may physically damage the organism by impairing locomotion, feeding, respiration, etc. Both types of damage are considered in the present study.

Short-term toxicity studies, designed to assess chemical damage from seawater soluble components of the oil, were conducted on a number of species. Adult forms of benthic crustaceans examined (an amphipod and a cumacean) are rather tolerant of high concentrations of oil in seawater. A benthic bivalve also proved to be highly tolerant, although an initial avoidance response, whose magnitude and duration varied directly with the oil concentration, was evident. A benthic tunicate was adversely affected by the oil and succumbed in moderate concentrations. Only one planktonic species was investigated. This small locally abundant medusa

was adversely affected by fairly low concentrations of oil in seawater, and proved to be the least tolerant of all the species examined.

It must be emphasized that these results are based on short-term lethal studies and that even those species that are classed as tolerant of crude oil by this criterion may still be adversely affected and eventually succumb over a more extended period. For this reason more sensitive physiological criteria are required for detecting sublethal deleterious effects. Metabolic rate is a sensitive indicator of an organism's general physiological state. We have investigated the effect of acute exposure to seawater soluble components of Arctic crude oils upon the metabolism of the aphipod Onisimus affinis. Neither Atkinson Point nor Norman Wells crude had any significant effect on respiration at low or moderate concentrations. At concentrations of 1000 ppm. and 5000 ppm. Atkinson Point oil increases respiration 25% and 30% respectively. Similar concentrations of Norman Wells oil increase respiration by 25% and 38%, respectively. There are indications that at still higher concentrations of both oils the rate begins to decline.

It appears that adult benthic forms such as Onisimus will not be susceptible to damage by direct toxic effects of seawater soluble components of crude oil at the concentrations likely to be encountered in the natural environment. Physical effects, resulting from direct contact with oil masses are likely to prove more damaging. This is particularly true for those species, such as Onisimus, which although normally benthic, congregate in large numbers in the vicinity of the under-ice surface during the winter. It is in this area that pockets of spilled oil are likely to accumulate. A series of experiments was conducted to assess the ability of Onisimus to recover following a brief immersion in several crude oils and return to clean seawater. Mortality occurs gradually over an extended period, and the "condition" of the animals (measured quantitatively by an activity index) declines progressively. Four weeks after exposure less than 20% of the animals are capable of swimming, and in most of these swimming is erratic and abnormal. Very few, if any, of the animals appear to recover fully.

In view of the fact that physical contact with crude oil may present the greatest potential danger

to certain species it is of interest to determine the behavioural response of marine organisms to oil masses present in their immediate vicinity. To permit comparison of results we have defined an affinity coefficient that indicates the degree of attraction or repulsion of a given species for a particular crude oil. Of three Arctic crustaceans studies two amphipod species exhibit a significant avoidance response to oil masses, the type of oil markedly influencing the degree of avoidance. The isopod Mesidotea appeared to be quite indifferent to the presence of either Atkinson Point crude or Venezuela crude. In general, Arctic crude oils were more repellent than Venezuela crude.

Initial indications are that adults of a number of benthic species are fairly tolerant of short-term exposure to moderate concentrations of crude oils. Since an intertidal community is absent in most areas of the Arctic because of ice scouring, oiling of beaches will not result in the massive smothering mortalities common in temperate regions. Species most likely to be heavily damaged in the event of an oil spill include planktonic forms and those benthic species that migrate to the undersurface of the ice in winter. In the case of the latter animals contact with crude oil probably poses the greatest potential threat. However, indications are that certain species actively avoid spilled oil. The degree of avoidance varies with both the species and the type of oil involved. A review of other potential adverse effects of oil pollution on marine invertebrate communities in the Arctic is presented to illustrate the magnitude of the problems that remain.

INTRODUCTION

It has been stated with good reason that oil pollution is the most serious and immediate threat facing the Arctic ecosystem (Dunbar, 1971). This is particularly true for the Arctic marine ecosystem. Recent developments in both the Canadian north and in Alaska, related to the extraction and transport of crude oil, have rudely awakened us to the fact that our present understanding of both short- and long-term consequences of such activities upon Arctic marine life is woefully inadequate. As Dunbar (1971) has so aptly put it "we have been caught in a state of scientific near nudity in the particular respect in which we now so urgently need protective covering; namely knowledge of what the proposed developments will do to the environment, in precise terms, and knowledge of what should be done to conserve and protect." That we know little or nothing about even the natural physiological ecology of most Arctic marine species is further cause for concern. What little information that we do have strongly suggests that simple extrapolations from studies conducted in temperate regions are of questionable significance in the Arctic context. Arctic species exhibit a variety of physiological and ecological adaptations, about which we know very little, that permit them to thrive in a unique and very demanding environment.

That the ecological impact of oil pollution in the north is likely to be particularly severe can be deduced from the following generalizations about Arctic marine ecosystems:

1. Limited numbers of species and possible ecosystem instability (Dunbar, 1968)

2. Slow growth, extended life cycles and longer reproductive periodicity (Chia, 1970)

3. Slow biodegradation and dispersion of oil at low temperature.

These particular features suggest a long drawn-out process of recovery once damage occurs. However, there still remains the important question of just how susceptible the various components of the ecosystem are to significant damage in the first place. It is to this question that the present study primarily addresses

itself. The study was carried out in conjunction with a marine invertebrate ecological physiology program at the Arctic Biological Station and thus it tends to be primarily physiologically oriented. It represents an attempt to gain some insight into potentially harmful interactions between some of the dominant marine invertebrates of the Mackenzie delta region and Arctic crude oils.

Studies conducted in temperate and tropical regions very clearly indicate that different species vary markedly in their tolerance of oil pollution. We were therefore interested in determining the relative sensitivity to Arctic crude oil of various ecologically important marine invertebrates present in the study area. Crude oil may directly damage organisms in two distinct ways. Certain seawater soluble components may exert a lethal toxic effect by chemically interfering with cell or tissue functions. On the other hand, the whole oil itself, by virtue of its high viscosity and hydrophobic nature, may physically impede the normal functioning of organisms and thus impair locomotion, feeding, respiration, etc. Both toxicity by soluble components and impairment of function by physical contact with whole oil are considered in the present study.

The likelihood of high mortalities resulting from contact with oil masses raises the question of just how organisms react to the presence of discrete masses of oil in their immediate vicinity. Do they generally tend to avoid it? Are they attracted to it? Or do they react as if it didn't exist and only blunder into it on a chance basis? Very little reliable information is available regarding the behavioral response of marine invertebrates to crude oil. In view of the paucity of data, and the potential ecological significance of an avoidance or attraction response to certain of the species being studied we conducted a series of tests to determine the affinity of several marine invertebrates for a number of different crude oils.

Useful as short-term toxicity studies are for determining the relative sensitivity of various species and the relative toxicity of various crude oils, they provide little realistic information about the actual levels of pollutants that may ultimately prove detrimental to an ecosystem. At best, they permit an estimate of gross biological effects that result from massive concentrations of oil, usually far in excess of levels

likely to be encountered in the natural habitat, except perhaps in very restricted areas. Increasing attention is being directed to more subtle sublethal effects, which though more difficult to assess, nevertheless, yield a much more realistic estimate of the potential impact of oil pollution on marine communities. The rate of oxygen consumption of an organism is frequently utilized as a useful measure of general metabolic activity and provides a sensitive monitor of subtle changes in an animal's physiological state when subjected to an environmental stress. In the present study we examine the acute influence of a number of crude oils on the metabolic rate of a marine invertebrate that on the basis of short-term toxicity texts is considered resistant to high concentrations of crude oil.

RESUME OF CURRENT STATE OF KNOWLEDGE

1. The Nature of the Threat of Oil Pollution to Marine Organisms

Studies on the impact of petroleum pollution upon marine ecosystems often yield conflicting or inconclusive results that make it very difficult to establish general principles. That this is so is largely due to the very complex nature of the pollutant, both in its chemical and physical characteristics, and to the diverse physiological responses of different types of organisms to various components of the oil. After years of study, both in the field and in the laboratory, by many investigators, our knowledge of the short-term effects of relatively massive quantities of oil and oil products is extensive, though by no means complete. Concerning the more subtle, long-term effects of relatively low concentrations of oil we know virtually nothing. It is to this latter area that much present day research is directed.

Petroleum pollutants generally fall into two very broad categories. Non-persistent pollutants include the lighter refined components such as gasoline, kerosene, diesel oil, etc. These are volatile components that have generally been found to evaporate rather quickly following a spill. Persistent pollutants such as crude oil, bunker fuel etc. have a large bulk of essentially non-volatile components that may remain in the environment

for an indefinite period of time. In the absence of far northern refining facilities it is the latter type of pollutants that present the most serious threat to Arctic ecosystems. It is generally assumed that the toxicity of refined products is greater than that of crude oil; however, Butler and Berkes (1972) after an extensive review of the literature suggest that the available data does not warrant such a conclusion. Shelton (1971) maintains that apart from the light refined components, petroleum products are not highly toxic. This would also appear to be the general conclusion to be drawn from the extensive series of studies that followed upon the Torrey Canyon incident (Smith, 1970). However, toxic effects of crude oils have been widely reported (see review by Butler and Berkes, 1972), and Allen (1971) found that crude oils and heavier bunker oils have a significantly greater inhibiting effect upon cleavage of sea urchin eggs than do more highly refined petroleum products. Much of the confusion, no doubt, arises from the fact that different types of crude oil vary quite extensively in their toxic effects as Ottway (1971) has clearly shown for several species of intertidal organisms. Kuhnhold (1970) reports a similar phenomenon with regard to crude oil toxicity to herring eggs.

Blumer (1970) in summarizing some of the potential biological effects of oil pollution suggested that adverse effects upon organisms can be conveniently grouped into the following categories:

(a) Direct kill of organisms by physical effects such as smothering.

(b) Direct kill of organisms by contact toxicity i.e. by components that have only limited solubility in seawater.

(c) Direct kill or organisms by seawater soluble compounds.

(d) Destruction of sensitive larvae and eggs.

(e) Destruction of food sources.

(f) Sublethal effects resulting in reduced tolerance to normal stresses.

(g) Incorporation and possible concentration of carcinogens and other potentially toxic compounds into food chains.

(h) Interference with subtle integrative mechanisms of populations or communities.

2. Effects of Direct Contact with Crude Oil

As has been pointed out, adverse effects of contact with crude oil may arise from two distinct causes; a purely physical impairment of function as a consequence of the high viscosity of the oil, and a chemical disruption of physiological functions resulting from the presence of low solubility components in the oil. In practice it is difficult to distinguish between these two effects. In many cases it is likely that both factors acting in concert contribute to the death of the organism.

The purely physical effect may involve interference with respiratory exchange by clogging gill chambers, formation of a film over gill surfaces or immobilization of appendages essential for ventilation. Simple immobilization of the organism in the oil mass may lead to ultimate death. Such physical smothering effects are a particular hazard to intertidal communities.

Little is known about the nature of contact toxicity. Many studies have shown high mortalities of various species immersed in crude oil for short periods and then washed and transferred to clean seawater (Crapp, 1969; Nelson-smith, 1968; Mironov, 1967). Many of the species that showed considerable susceptibility to this treatment were molluscs, supposedly equipped with a protective shell. In most such studies it is impossible to determine whether the animals are succumbing from physical or chemical causes. Crapp (1971), however, found that immersion of several species of molluscs in fresh crude oil led to high mortality, while similar exposure to weathered crude oil residue did not lead to significant mortality. This appears to rule out a physical cause, and indicates that a toxic component is responsible for the mortality and that it is a light volatile component that is lost on weathering. The practical significance of this is that freshly spilled crude oil washed ashore rapidly is likely to do considerably more biological damage than is oil that has been weathering at sea for some time before coming ashore. This is believed to be one of the reasons why in the Torrey Canyon oil spill the crude oil itself had only a limited effect on shore invertebrates (Smith, 1970).

3. Toxicity of Soluble Components of Crude Oil

Crude oil is a complex mixture of a diverse assortment of hydrocarbons, many sulfur and nitrogen containing compounds and a wide variety of inorganic compounds. The water solubility of many of the hydrocarbon constituents appears to be relatively low. However, certain phenolic compounds, straight chain hydrocarbons up to C_8 and a number of the aromatic compounds are quite soluble (Dean, 1968; McKee, 1956; Hodgman et al., 1960; Wardley Smith, 1968).

The different components of the crude oil differ markedly in their degree of toxicity. Much of the toxicity appears to be associated with certain of the aromatic fractions boiling below 149°C (Ottway, 1971). The numerous studies conducted on the toxicity of various isolated components of crude oil have been reviewed by Butler and Berkes (1972). A number of generalizations may be drawn from these studies. Toxicity generally increases along the series paraffins, naphthenes, olefins and aromatics. Within a given series smaller molecules tend to be more toxic than large; thus octane and decane are very toxic, while dodecane and higher paraffins are virtually non-toxic (Van Overbeek and Blondeau, 1954). Unsaturated hydrocarbons naphthenic acids and compounds containing aromatic groups contribute to the total toxicity. Russian work, quoted by Galtsoff (1936) indicates that hexhydrobenzoic acid is one of the highly toxic components of Baku crude. Toxicity studies on various other refined constituents of crude oil have been reviewed by Nelson-Smith (1971). Considerably more work is required on the lethal and sublethal effects of other components of crude oils, particularly those components that are of a persistent nature.

The very complex and highly variable composition of crude oils accounts for the considerable differences in toxicity observed for different crude oils. The complexity of composition and wide variability in the solubilities of the different components also accounts for the difficulty encountered in conducting meaningful toxicity tests; one is never very sure of just how much of which types of toxic compounds are actually getting to the organism.

4. Variability of Species Sensitivity to Oil Pollution

Even a cursory examination of the considerable literature on the biological effects of oil pollution reveals a marked difference in the ability of different types of animals to tolerate the presence of oil in their environment. Even allowing for different types of oil, different preparative and experimental techniques one is, nevertheless, forced to the conclusion that certain species can thrive in incredibly high concentrations of oil while other species are killed or severely stressed by even a hint of petroleum in their environment. The precise reason for this remarkable difference in tolerance is not at present known, largely due to the fact that we have no very clear idea of which components, and what mechanisms of toxicity, are responsible for irreversible damage to organisms exposed to a given crude oil. The following is a brief summary of a number of reports substantiating the above conclusion. It is by no means an exhaustive review of the subject.

Data on sensitivities of various species to oil pollution has been derived from two different sources; field observations following accidental or controlled oil spills and laboratory toxicity studies conducted under standardized environmental conditions. The former are usually difficult to interpret because of the lack of controls and the potential involvement of a wide range of uncontrolable environmental variables that may modify the overall effect of the oil.

Studies by Mironov (1970) suggest that zooplankton may be particularly susceptible to crude oil. Although most species examined (mostly copepods) tolerated 1 ppm. almost all species experienced high mortality within 24 hours when exposed to 100 ppm. oil. There is little firm field data regarding zooplankton mortality in the vicinity of oil spills largely because of difficulties in assessing effects and a lack of adequate baseline information for the areas in question. According to Smith (1970) there appeared to be "little detected damage suffered by planctonic organisms in the western English Channel following the release of oil from the Torrey Canyon". However, larval stages of a wide range of marine invertebrates that at times constitute a substantial portion of the zooplankton community have in general been found to be very sensitive to petroleum (Wells, 1972; Chia, 1973; Mironov, 1969).

Considerably more information is available regarding the effect of oil on benthic invertebrates, although much of it appears contradictory. Intertidal organisms have received the most extensive studies largely because effects in the field are usually obvious and easy to assess and because these species inhabit areas that frequently bear the brunt of oil pollution incidents.

Sea anemones of the genus anthropleura are reported to be very resistant to oil pollution, even surviving heavy coating and smothering (Foster et al., 1971). In fact North et al. (1964) report that not only did Anthropleura xanthogrammia suffer few ill effects from the Tampico Maru oil spill but that it occurs commonly in the effluent pools of a California refinery. However, other species of anemones appear to be very sensitive to oil (Manwell and Baker, 1967).

Molluscs too vary considerably in their response to oil. Thus successive immersion of oysters in oil caused little mortality, although as pointed out earlier sublethal deleterious effects are evident (Chipman and Galtsoff, 1949). Clams, Mercenaria mercenaria, are reported to live in bays in Rhode Island the bottoms of which are literally "paved with oil" (Hawkes, 1961). The mussel Mytilus galloprovincialis is able to survive and function in high concentrations of oil although abnormal behavioural effects are evident at very high concentrations (Alykrinskaya, 1966). In support of this, Foster et al. (1971) reports that no significant mortality occurred among mussels, chitons and limpets following the Santa Barbara oil spill. However, not all bivalves are as tolerant of oil pollution as are these species. Large number of pismo clams and abalones were killed by the oil released in the Tampico Maru spill (North et al., 1964). A fuel oil spill on the U.S. west coast resulted in the death of an estimated 300,000 razor clams in less than a week (Tegelberg, 1964). Gastropod molluscs are also susceptible to oil. The winkles Littorina littorea and L. obtusata experience high mortalities when fresh crude oil was poured over them and then rinsed off (Crapp, 1969). Similar treatment resulted in high mortality in the limpet Patella vulgata (Nelson-Smith, 1968) and to the gastropods Bittium reticulatum, Rissoa euxinica, and Gibbula divaricata (Mironov, 1967).

Results for crustaceans are similarly variable,

depending upon the oil type and nature of the exposure to the oil. Field experiments with barnacles suggest no obvious ill effects from exposure to crude oil (Crapp, 1969). However, the barnacle Chthamalus fissus suffered high mortality from smothering and heavily oiled goose-neck barnacles also died in the wake of the Santa Barbara oil spill (Foster et al., 1971). That more than simple smothering may be involved is suggested by studies of Chipman and Galtsoff (1949) who found that some species of barnacles are killed by as little as 2% crude oil in less than three days. Lobsters and crabs suffered heavy mortality following the Tampico Maru spill (North et al., 1964). Large numbers of lobsters and other crustaceans were killed and washed up on the shore after a fuel oil spill in Buzzards Bay (Blumer et al. , 1971). However, lobsters were not significantly affected by the Bunker C oil spilled from the Arrow in Nova Scotia (Scarratt, 1970). Large numbers of sand amphipods (Orchestoidea sp.) were oiled and killed in the Santa Barbara spill (Foster et al., 1970). Another sensitive amphipod rapidly succumbed in the presence of oil in the Buzzards Bay spill and could be used as an indicator of pollution (Blumer et al., 1971). In marked contrast, the crab Carcinus maenas, along with the sea squirt Ciona intestinalis and the jellyfish Aurelia aurita are common in heavily oiled dock areas around Swansea (Naylor, 1965). Some coral species are sensitive to oil (Lewis, 1971) as are sea stars and sea urchins (Nelson-Smith, 1970). Marine worms, from the few studies that have been reported appear to be very tolerant of oil pollution (Foster et al., 1971; Blumer et al., 1971).

On the basis of the above data it is difficult to draw hard and fast generalizations regarding the oil tolerance of various classes of invertebrates. Difficulties in making comparisons arise from the fact that differences in the tolerance of species are obscured by differences in the oils involved, differences in method of application of the oils, differences in the degree of weathering of the oil, in addition to differences in a variety of other environmental parameters.

The overall ecological effect of the differential sensitivity of species to oil is that after a major pollution incident sensitive species are rapidly eliminated. More tolerant species, confronted with less competition and/or predation, may undergo a virtual population explosion. Such an effect was observed following the Buzzards Bay oil spill. The marine worm

Capitella capitata which usually occurs in the area in small numbers became very abundant following the spill and flourished in all but the most heavily polluted areas (Blumer et al., 1971). Nelson-Smith (1971) suggests that chronic pollution (low-level, long-term) also leads to a reduction in diversity of species in an area. This would in general tend to increase ecological instability.

5. Sublethal Effects of Crude Oil

For many years short-term toxicity tests have been the favorite research tool of pollution biologists interested in gaining an insight into the effect of various pollutants upon aquatic organisms. Mortality is easy to measure and obviously detrimental to the individual concerned. Useful as such studies have been in outlining the nature and general scope of pollution damage they can never be considered as more than crude first approximations in our understanding of the problem. Of more importance in the long run are those subtle noxious effects that while they don't kill individuals outright tend nevertheless to the extermination of the population in the course of one or several generations. Such effects are often difficult to detect and it is only now that we are gaining awareness of the many insidious forms such sublethal effects can take. For most pollutants, including crude oil, we have no reliable estimates of the critical concentrations of the pollutants that induce ecologically significant sublethal effects.

A number of studies have demonstrated that crude oil can adversely affect the nutrition of marine organisms. Chipman and Galtsoff (1949) reported that successive immersions of oysters in crude oil caused little mortality although storage of glycogen appeared to be inhibited. Later work revealed that soluble components of the oil have an anaesthetic effect upon gill cilia leading to reduction in pumping with a consequent reduction in feeding and storage of reserves (Galtsoff, 1964). Similarly, Kuwait crude oil has a depressant effect on cirral activity in barnacles leading to a reduction in feeding and an inhibition in growth (Smith, 1970). The coral, Madracia asperula, reduced its feeding significantly following 24 hours exposure to as little as 10 ppm crude oil. No recovery occurred upon return to clean seawater (Lewis, 1971). One nutritional aspect of oil pollution that has not

been examined in any great detail is the effect of ingestion of crude oil upon marine invertebrates. Oil droplets are readily ingested by copepods (Conover, 1971), periwinkles and sea urchins (Zitko and Carson, 1970). After the Torrey Canyon oil spill as much as 50% oil was detected in the feces of intertidal molluscs (Smith, 1970). It is unlikely that the oil passes through the animal unchanged. What components are absorbed from the oil and what effect the presence of oil has upon digestive processes is now known.

Little is known concerning the effects of petroleum products upon the normal activity and behavior of marine organisms. Hargrave and Newcombe (1973) observed an increased rate of crawling of the gastropod Littorina in the presence of seawater extracts of Bunker C. Whether or not this represents an escape response is unclear. Depressed gill ventilation in oysters and cirral beating in barnacles as a result of exposure to crude oil have been alluded to above. Changes in locomotory activity caused by crude oil could have important consequences in terms of ability to feed or to escape from predators. Whether petroleum products have an adverse effect upon behavioral responses to normal environmental stimuli is not known. Whether oil interferes with the function of chemoreceptors is not known. Many marine invertebrates rely on chemo receptors for finding food. Blumer (1970) suggested that petroleum products may attract lobsters away from their natural food. It is conceivable that certain components of crude oils may mimic natural "messenger" compounds and evoke inappropriate responses in organisms.

One of the most subtle and difficult to detect sublethal effects of pollutants is that they may reduce the resistance of organisms to normal environmental stresses. These may involve physical stresses such as temperature, salinity or other pollutants; or biological stresses such as predation, competition, disease and parasites. Synergistic effects of this type have been demonstrated for pollutants such as mercury (Vernberg and Vernberg, 1972). Little is known about the potential involvement of oil in such synergisms. Sublethal pollutant stress would be particularly critical to populations that are near the limits of the species range.

6. Affinity of Marine Organisms for Oil Masses

As pointed out earlier very little reliable

information is available regarding the behavioral responses of marine organisms to crude oil masses in their immediate vicinity. A number of field observations are suggestive but of uncertain significance. Reports quoted by Nelson-Smith (1971) indicate that some fish species tend to avoid spilled oil drifting on the sea surface. Blumer (1970) notes that certain purified hydrocarbons derived from kerosene attract lobsters, and he suggests that the massive mortality experienced by this species following the Buzzards Bay oil spill may in part be attributable to the fact that animals were attracted away from their normal food in the direction of the spill. Wilder (1970) observed that lobsters readily consume fish heavily contaminated with Bunker C oil. Unfortunately no observations were made to determine if the contaminated food was eaten more readily than uncontaminated food. In connection with the Arrow spill of Bunker C Thomas (1970) reported that periwinkles, Littorina sp. appeared to migrate from heavily oiled areas to adjacent clean areas. Furthermore, he noted that clams, Mya arenaria, generally moved out of oil polluted burrows. It is not clear whether this represented an aversion to the oil or a simple attempt to escape suffocation in the burrows. Definitive laboratory studies on the behavioral responses of marine invertebrates to petroleum and petroleum products are clearly lacking.

STUDY AREA

Most of the animals used in these studies were collected in the Eskimo Lakes, an interconnected chain of lakes that form a relatively shallow, estuarine extension of Liverpool Bay adjacent to the MacKenzie Delta. Isopods employed in the affinity tests were collected in shallow waters along the north western shore of Liverpool Bay. Experience in temperate regions suggests that such shallow, semi-enclosed coastal waters are likely to experience the most severe impact of an oil pollution incident. In such confined areas spilled petroleum tends to remain concentrated and to persist for extended periods, in contrast to the situation that occurs in the open sea where spilled oil is usually dispersed over a fairly wide area very rapidly.

MATERIALS AND METHODS

1. Toxicity of Seawater Soluble Components

Animals used in these studies were collected in the Eskimo Lakes and held in a refrigerated aquarium at approximately normal environmental temperature and salinity at the field station until used. The species tested included the amphipod Onisimus affinis, the tunicate Rhizomolgula globularis, the bivalve mollusc Yoldiella intermedia, the coelenterate medusa Halitholus cirratus, and the cumacean Brachydiastylis resima. Only adults of each species were used.

Fresh Atkinson Point crude oil was employed in the tests. Seawater extracts of the oil were prepared as follows: 100 ml. of the crude oil was placed in a large stoppered flask with one liter of filtered natural seawater of salinity 17-18 ppt. The contents of the flask were stirred vigorously on a magnetic stirrer for 6 hours at room temperature at a standard speed (the vortex formed extended approximately one-quarter of the way down into the solution). The contents of the flask were then transferred to a separatory funnel and allowed to stand for two hours while the two phases separated. The seawater/oil extract was drawn off and filtered through Whatman no. 1 filter paper. This stock solution was then used to prepare dilution of 10, 100, 1000, 5000 and 10,000 ppm. These dilutions are not absolute measures of the concentrations of components of the crude oil, they merely indicate the relative concentrations of those components that are reasonably soluble in seawater. During the test the animals were held in plexiglass chambers of approximately 500 ml. capacity immersed in a constant temperature bath held at 7-8^{o}C. A multi-channel peristaltic pump delivered the oil/seawater mixtures from one gallon polyethylene reservoirs to each of the chambers at a rate of approximately 100 ml./hr. One of the chambers was designated a control chamber and received a similar flow of filtered natural seawater. Excess water from each of the chambers overflowed to a drain. The seawater/oil solutions were prepared fresh as necessary to replenish the reservoirs.

Sixteen to 25 (depending upon the species) healthy animals were placed in each of the experimental chambers. The chambers were checked at regular intervals

for 96 hours and each time the number of active animals was tabulated. The definition of "active" varied with the species. For the crustaceans Onisimus and Brachydiastylis an active animal was considered to be one exhibiting swimming activity and/or coordinated appendage movement. For the mollusc Yoldiella the criterion of activity was the opening of the valves accompanied by an extension and sweeping motion of the foot. For the tunicate rhizomolgula an animal was considered active if the body was fully distended and with the incurrent and excurrent siphons extended and open yet capable of rapid retraction when disturbed. The medusa Halitholus was considered active if it exhibited regular, coordinated pulsations of the bell. At the end of 96 hours the number of animals still alive was determined. A Crustacean not exhibiting appendage movement was considered dead. Medusa were considered dead if they exhibited no bell pulsation or had lost their normal turgid form. Rhizomolgula were considered dead if the siphons were retracted and the body was flaccid. It was not possible to define a readily detectable indicator of death for the bivalve Yoldiella because of the presence of the protective shell. However, as more than 90% of the animals were clearly alive and active after 96 hours in even the highest oil concentration tested this did not present a problem.

2. Contact Toxicity

Animals for these studies were collected in the Eskimo Lakes and maintained in the circulating seawater system at Ste. Anne de Bellevue until used; holding temperature 0°-2°C, salinity 17-18 ppt. Tests were conducted on the amphipod Onisimus affinis which had previously been shown to be rather resistant to oil/seawater solutions. Three crude oils were used; Atkinson Point crude, Norman Wells crude and Venezuela light crude.

Groups of 15 adult animals were immersed in about 10 ml. of one of the crude oils for either 30 seconds (series 1) or two minutes (series 2). The animals were then poured onto a fibreglass screen and rinsed with approximately 1 liter of clean seawater. The animals were then transferred to polyethylene beakers containing 400 ml. of seawater that was aerated continuously. A control group, treated similarly except that it wasn't exposed to oil, was also prepared. The beakers

were held in a constant temperature room at 0°C. After one hour and then at approximately daily intervals for 30 days the animals were transferred to a shallow glass tray and scored for activity according to the following numerical scale:

3 - animal exhibits spontaneous swimming activity within a five minute observation period. Preliminary tests had demonstrated that in a group of normal animals all of the individuals generally exhibited spontaneous swimming within two minutes.

2 - animals exhibit coordinated limb movement including rhythmic beating of the pleopods, however, they do not swim during the five minute observation period.

1 - animals exhibit slight, irregular, uncoordinated limb movement.
pleopod beat irregular if present.

0 - no detectable appendage movement, animal dead.

After each observation period the animals were returned to the beakers which had been refilled with fresh seawater.

The activity scores obtained during each observation period were added to give an estimate of the locomotory capability of the group at that particular time. The maximum value for any given group is 45. The locomotory capability at each observation was expressed as a percentage of this maximum value. In addition, for each observation, the percentage of animals swimming spontaneously and the percentage of dead animals was calculated.

3. Affinity for Crude Oils

Animals used in these studies were collected in the Eskimo Lakes and Liverpool Bay and were maintained in the circulating seawater system at the Ste. Anne de Bellevue laboratory prior to use; holding temperature 0-2°C, salinity 17-18 ppt. The three species tested included the amphipods Onisimus affinis and Gammarus oceanicus and the isopod Mesidotea entomon. Three types

of crude oil were tested; Atkinson Point crude, Norman Wells crude and Venezuela light crude.

Test chambers consisted of shallow polyethylene trays subdivided into four equal zones (designated A, B, C and D) by lines inscribed on the bottom of the tray. For the smaller Onisimus, the chamber measured 40 cm. long, 8 cm. wide and 4 cm. deep. and was divided linearly into 4 zones. For the larger Gammarus and Mesidotea a chamber measuring 33 cm. long by 28 cm. wide by 5 cm. deep divided into quarters was used. Preliminary tests revealed that significant numbers of Onisimus became trapped by surface tension and were thus impeded in their movement within the chamber. This problem was finally overcome by completely filling the chamber with seawater and placing a clear plexiglass sheet on the water surface to eliminate the air/water interface. The two larger species were unaffected by surface tension, so for these, the water surface remained exposed. The experimental chambers were partially immersed in an ice bath to hold the temperature between 2^{o} and 4^{o}C during the course of a run. For all of the runs the trays were held in a constant orientation in the laboratory. In order to record the positions of the animals in the various zones at intervals, a camera was positioned over the chamber so that the entire interior of the chamber was included in the field of view. For each run, one zone was designated as an oiled zone and the other three as unoiled or control zones. To hold the oil, small squares of sponge measuring approximately 2cm x 2cm x 1cm were affixed to glass microscope slides with non-toxic silicone cement. Immediately prior to a run 1 ml of fresh crude oil of the desired type was placed on the sponge. As soon as the oil was absorbed the sponge was rinsed briefly in a large volume of seawater to remove excess oil; this rinsing was necessary to prevent the formation of oil slicks in the test chamber. The oiled sponge was then placed in the preselected oiled zone. Similar sponge squares that had not been treated with oil were placed in each of the control zones. The sponge squares were placed in the centre of each zone after the seawater and animals had been placed in the chamber, just prior to the commencement of the run. At the outset the animals were divided approximately equally among the zones.

To eliminate possible phototactic responses and to prevent the animals visually distinguishing between the dark colored oiled sponges and the paler unoiled sponges all runs were conducted in complete darkness,

except when observations were being made, at which time a brief period of illumination accompanied the photographing of the chamber.

Each run lasted for thirty minutes with the chamber being photographed at two minute intervals. Four runs with a single species and a single oil type constituted a series, with each of the four zones being successively designated as the oiled zone. At the end of each run the chamber was thoroughly rinsed and refilled with fresh seawater. A freshly oiled square of sponge was used for each run. The same group of animals was used for each run in a given series. At the end of each series the film was developed and the numbers of animals in each zone at each observation were counted from the negatives and tabulated.

For each run the numbers of animals in the oiled zone at each observation were added to give an observed frequency for the oiled zone designated $0'_z$ where z indicated the oiled zone for a given run. The numbers of animals in each of the three unoiled zones were similarly added and the totals from the three zones combined to yield an observed frequency for the unoiled control zones designated 0_c. For each run $0'_z$ and 0_c were combined to give the total number of counts for that run. Statistical probability dictates that in the absence of an attraction or repulsion response, for each run one quarter of the counts should occur in the oiled zone and three quarters in the unoiled zone. The total counts for a given run were divided accordingly to yield an expected frequency for the oiled zone, designated e'_z and an expected frequency for the unoiled zone designated e_c. Thus X^2 value was calculated from the observed and expected frequencies in oiled and unoiled zones for each run. The X^2 values for each of the four runs in a given series were added to yield a series X^2 value with four degrees of freedom (Spiegel, 1961). The probability of the observed frequency distribution in a given series being different from a random distribution was determined from X^2 tables.

In order to express the attraction or repulsion response in a clear and concise manner we have defined an affinity coefficient as follows:

$$A.C. = \frac{\Sigma 0'_z - \Sigma e'_z}{\Sigma e'_z} \times 100$$

where $\Sigma 0'_z$ is the sum of the observed counts in the oiled zone for each of the four runs in a series ($\Sigma 0'_z = 0'_a + 0'_b + 0'_c + 0'_d$) and $\Sigma e'_z$ is the sum of the expected counts in the oiled zone for each of the four runs in a given series ($\Sigma e'_z = e'_a + e'_b + e'_c + e'_d$).

An A.C. of zero indicates a neutral response and the animals are neither attracted nor repelled by the oil. An increasingly positive value indicates an increasing degree of attraction, to a maximum of 100 which indicates an absolute attraction to the oiled zone. Similarly, an increasingly negative value indicates an increasing degree of repulsion, to a minimum of -100 which indicates an absolute repulsion from the oiled zone. The significance of the A.C. for each series was determined from the X^2 values calculated as indicated above.

4. Effects of Oil on Respiration

Animals used in these studies were collected in the Eskimo Lakes and maintained in a refrigerated aquarium at approximately normal environmental temperature and salinity at the field station until used.

Oil in seawater solutions at dilutions of 10, 100, 1,000, 5,000 and 10,000 ppt. were prepared as described previously. Two types of crude oil were tested, Atkinson Point crude and Norman Wells crude. Although several species of invertebrates were examined only the data for Onisimus affinis is sufficiently complete at present to draw any meaningful conclusions; thus only the results for this particular species are presented.

Respiration rates were determined with a Gilson submarine respirometer using standard 15 ml. Warburg flasks. Four adult animals of a uniform size were placed in each flask. The medium consisted of 5 ml. of the appropriate oil/seawater solution or an equal volume of filtered natural seawater in the case of controls. Flasks were cleaned periodically with dichromate cleaning solution. The centre well of each flask was charged with 0.2 ml. of 20% KOH and a filter paper wick to absorb CO_2. Air was used as a gas phase and all runs were conducted at a temperature of 7.0°C. The shaking rate was 72 per minute. Flasks were equalibrated for one hour and readings taken at intervals (usually

30 minutes) for three to five hours. Upon completion of the run the animals were placed in vials and frozen. Later they were dried to constant weight at 70°c., cooled in a desiccator and weighed to the nearest 0.0001 gm.

Respiration rates were calculated from the slope of the line obtained by plotting cumulative oxygen consumption against time. Rates are expressed throughout at 10_2/mg dry wt/hr. Means and standard errors of the means were calculated and the significance of the difference of the respiration rate measured in each oil/seawater solutions from the control rate was determined by Students t test.

RESULTS

1. Short-Term Toxicity Tests

These tests are designed to indicate the susceptibility of animals to various concentrations of the seawater soluble components of the crude oil. All tests reported in this section were conducted with fresh Atkinson Point crude oil. As pointed out earlier, the concentrations referred to do not represent absolute quantities of the oil, but merely indicate the relative proportions of the seawater soluble components.

The amphipod Onisimus affinis appears to be relatively resistant to the crude oil, and over 90% of the animals remain alive and active throughout the duration of the test (Figure 1). It should be stressed that in this instance, the criterion of activity (coordinated limb movement) is not necessarily an indication that the animal's activity is entirely normal. We have some preliminary information that suggests that orientation and swimming activity may be impaired by the oil. We are presently taking a closer look at this particular aspect of the problem. However, even in the highest concentrations tested the animals were clearly alive and active even after 96 hours exposure to the pollutant.

The bivalve Yoldiella intermedia exhibits a rather interesting response to the presence of crude oil. For this species the criterion of activity was the opening of the shell, accompanied by extension and active sweeping of the foot (the normal mode of feeding

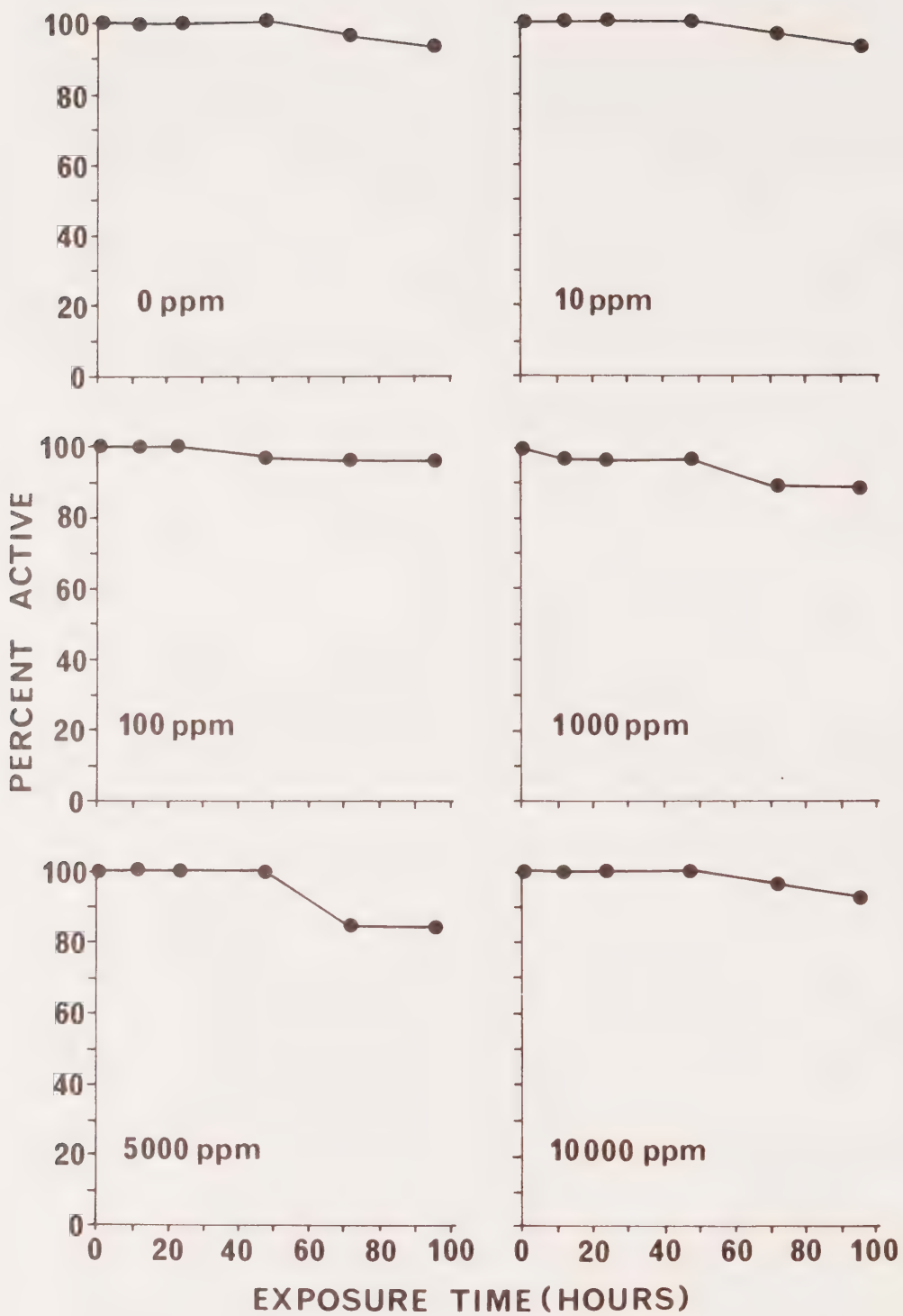

FIGURE 1. Activity of *Onisimus affinis* at intervals during 96 hour exposure to different concentrations of Atkinson Point crude oil.

in this animal). In normal unpolluted water more than 90% of the animals exhibited such activity at each observation period (Figure 2). When oil was present in the seawater a significant number of the animals responded by closing the shell and isolating themselves from the noxious environment. The percentage of animals that responded in this fashion increased as the concentration of oil increased. Thus in 10 ppm oil/seawater 30% of the animals ceased normal activity. In 100 ppm., 1,000 ppm., 5,000 ppm., and 10,000 ppm., approximately 35%, 50%, 60%, and 70% of the animals, respectively, exhibited the avoidance response. Thus the normal functioning of the animals is clearly adversely affected by the presence of oil in the surrounding seawater. However, the avoidance does not appear to represent a permanent response. After a period of time an increasing number of animals reopen and resume normal activity. The time that elapses before this resumption of activity increases with the concentration of the crude oil. Thus, in 10 ppm. the animals have returned to essentially full activity by 48 hours after initial exposure to the oil. Similarly, in 100 ppm. the animals require about 48 hours to resume activity. However, in 1,000 ppm. and 5,000 ppm. approximately 72 hours elapsed before a high level of activity was evident in the group. In the highest concentration tested, 10,000 ppm. activity only returned to normal after the lapse of approximately 106 hours. It is significant that even in this high a concentration most of the animals did eventually resume normal activity. It is clear from these results that Yoldiella is resistant over the short-term to fairly high levels of water soluble components of the pollutant.

In the case of the tunicate Rhizomolgula globularis, extension and opening of both incurrent and excurrent siphons and retraction of the siphons upon gentle stimulation, were used as criteria of normal activity.

Rhizomolgula is adversely affected by crude oil, but only at fairly high concentrations (Figure 3). Thus in 10 ppm. and 100 ppm oil/seawater all of the animals functioned normally for at least 72 hours. The decline in numbers active at 96 hours (75% active) parallels that exhibited by the controls and almost certainly represents an adverse response to confinement in the test chambers, quite independent of the presence of oil. The animals were not fed during the test period and it is conceivable that starvation effects may come into play after 72 hours. In 1,000 ppm. oil

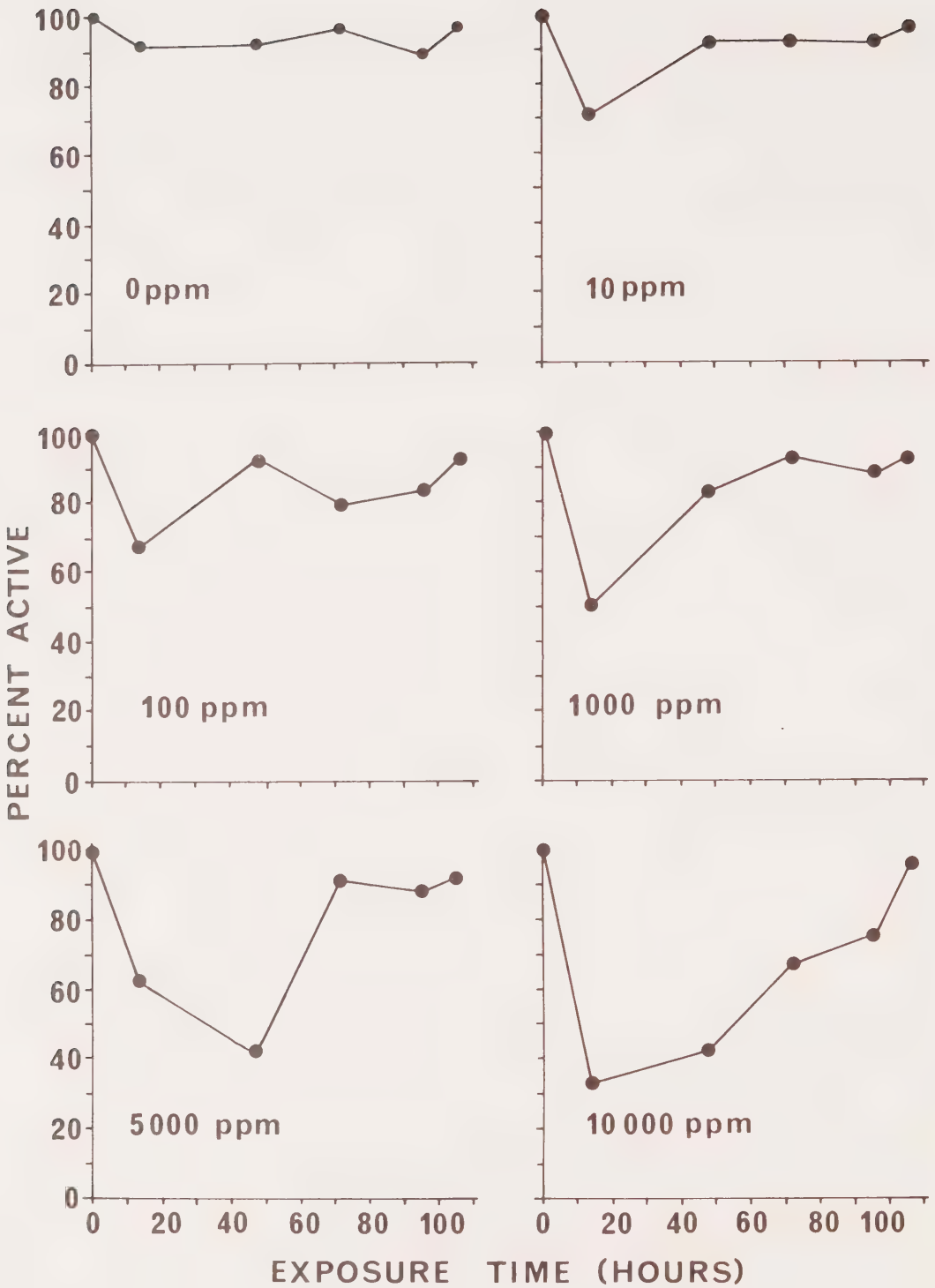

FIGURE 2. Activity of *Yoldiella intermedia* at intervals during 106 hour exposure to different concentrations of Atkinson Point crude oil.

the animals all function normally for at least 72 hours. By 96 hours only 45% of the animals were responding normally. Apparently at this concentration there is an adverse effect of the crude oil but it only manifests itself after an extended period. In contrast, in 5,000 ppm. and 1,000 ppm. the onset of impairment of functioning occurs more rapidly and by 48 hours significant numbers of animals have ceased pumping (20% and 25%, respectively). With time an increasing number of animals cease normal behaviour, so that after 96 hours only about 30% of the animals in 5,000 ppm. oil/seawater are actively pumping, while in 10,000 ppm. none of the animals exhibit normal behaviour.

Results for the medusa Halitholus cirratus are not quite as clear as for the other species examined and must be interpreted with caution. These planktonic animals are very sensitive to changes in their environment and are difficult to maintain in closed systems for extended periods. In the present study, bell pulsation, either spontaneous or when the water was agitated slightly was used as a criterion of activity. In the case of the control chamber, except during the initial observation, only 70% to 85% of the animals exhibited activity during the course of the run (Figure 4). As this number remained reasonably uniform for the full 96 hours it is possible that the initial rapid decline in numbers active reflects cessation of activity by a small group of animals damaged during collection, although every effort was made to select only normal healthy individuals. In the presence of increasing concentrations of oil an increasing number of animals became immobilized. In general, the animals became immobilized more rapidly as the oil concentration increased. None of the animals were active after 96 hours in either a 5,000 ppm. or 10,000 ppm. oil/seawater solution.

Crawling and/or coordinated limb movement was used as a criterion of normalcy for the cumacean Brachydiastylis resima. This species appears to be fairly resistant to crude oil components dissolved in seawater (Figure 5). Although significant mortalities occur in 100 ppm. and 1,000 ppm. oil/seawater solution these cannot be attributed to the presence of the oil, because a high degree of survival was observed in the 5,000 ppm. and 10,000 ppm. solutions.

Survival data for the five species after 96 hours exposure to various concentrations of the crude

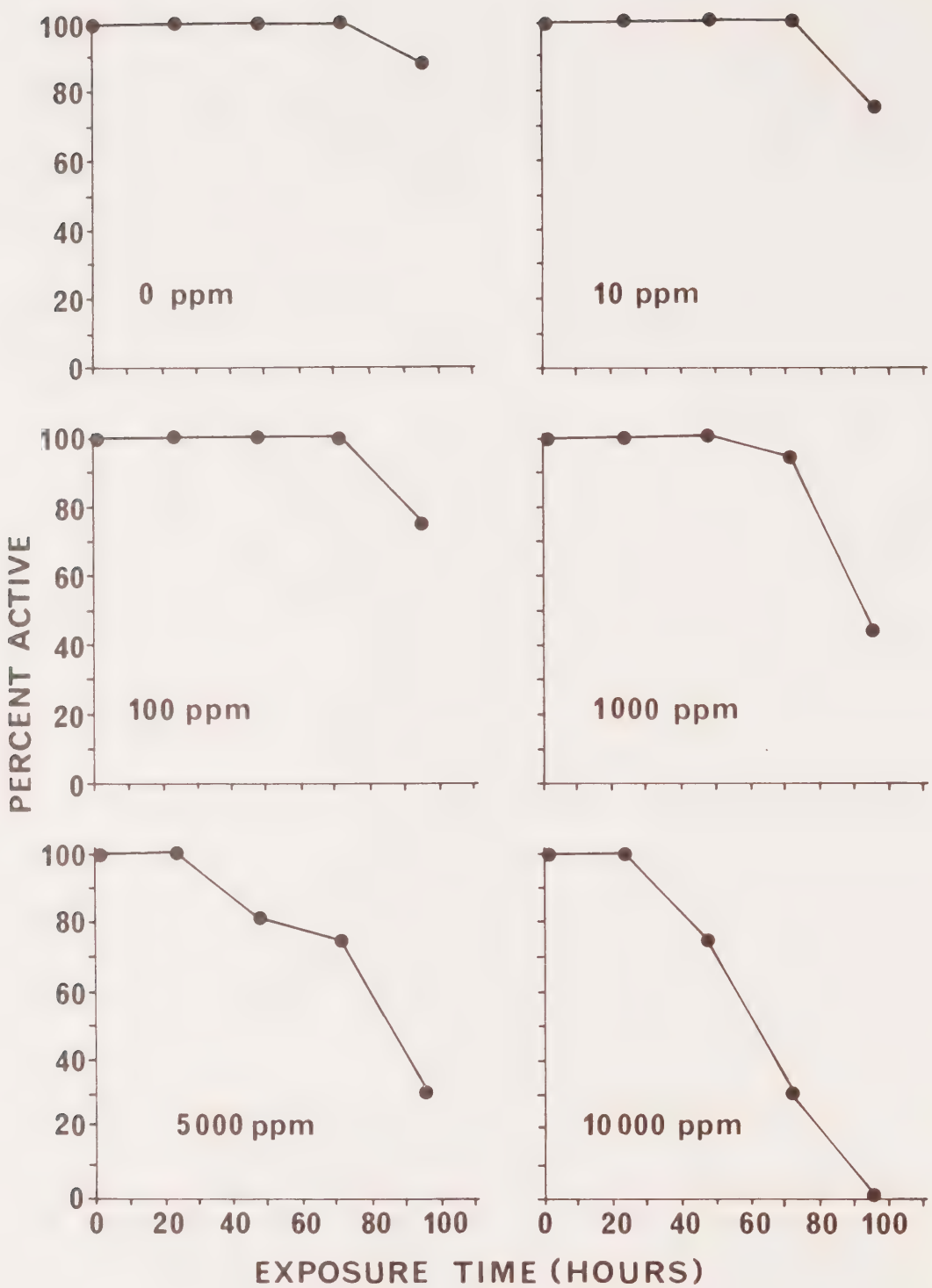

FIGURE 3. Activity of *Rhizomolgula globularis* at intervals during 96 hour exposure to different concentrations of Atkinson Point crude oil.

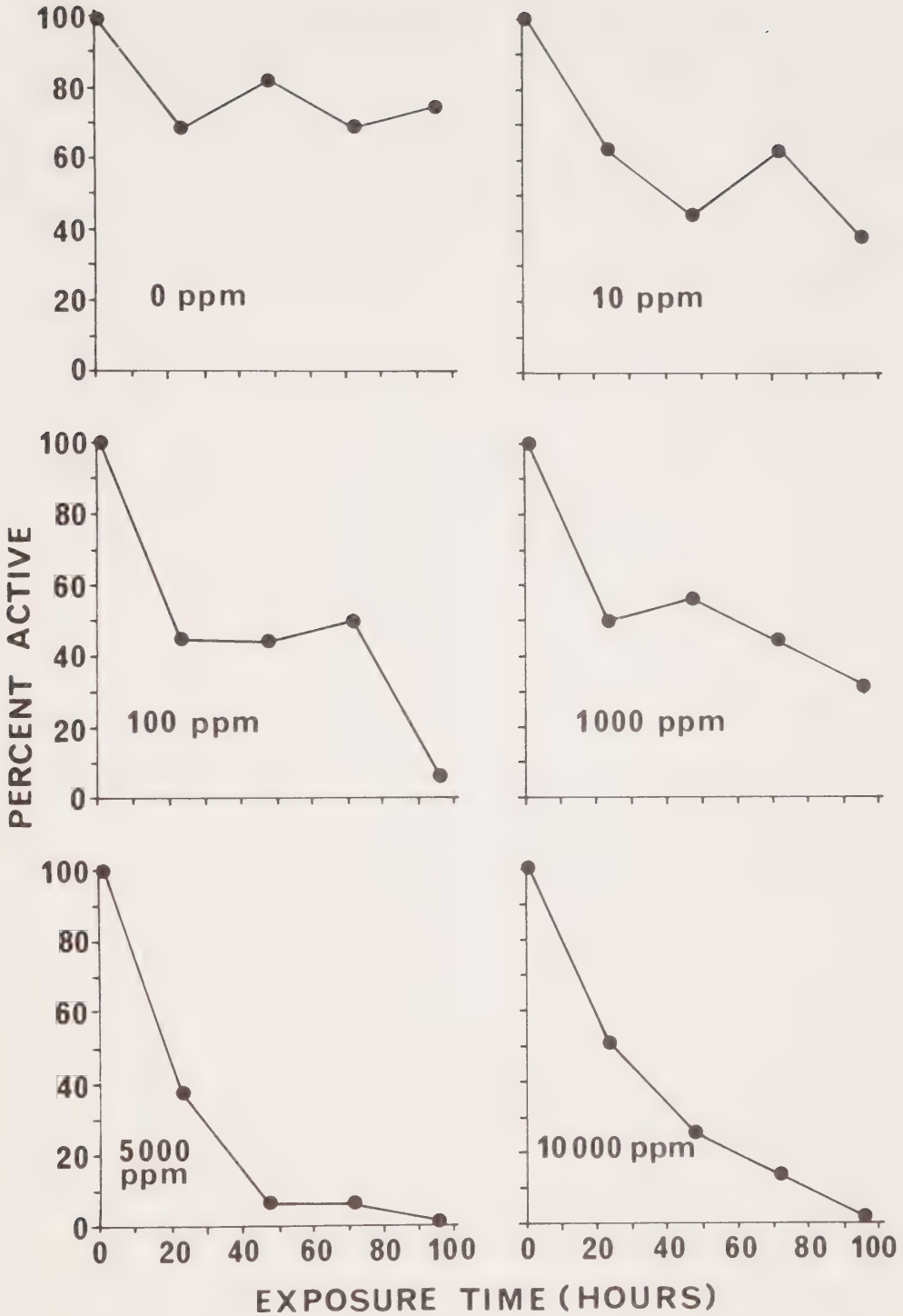

FIGURE 4. Activity of *Halitholus cirratus* at intervals during 96 hour exposure to different concentrations of Atkinson Point crude oil.

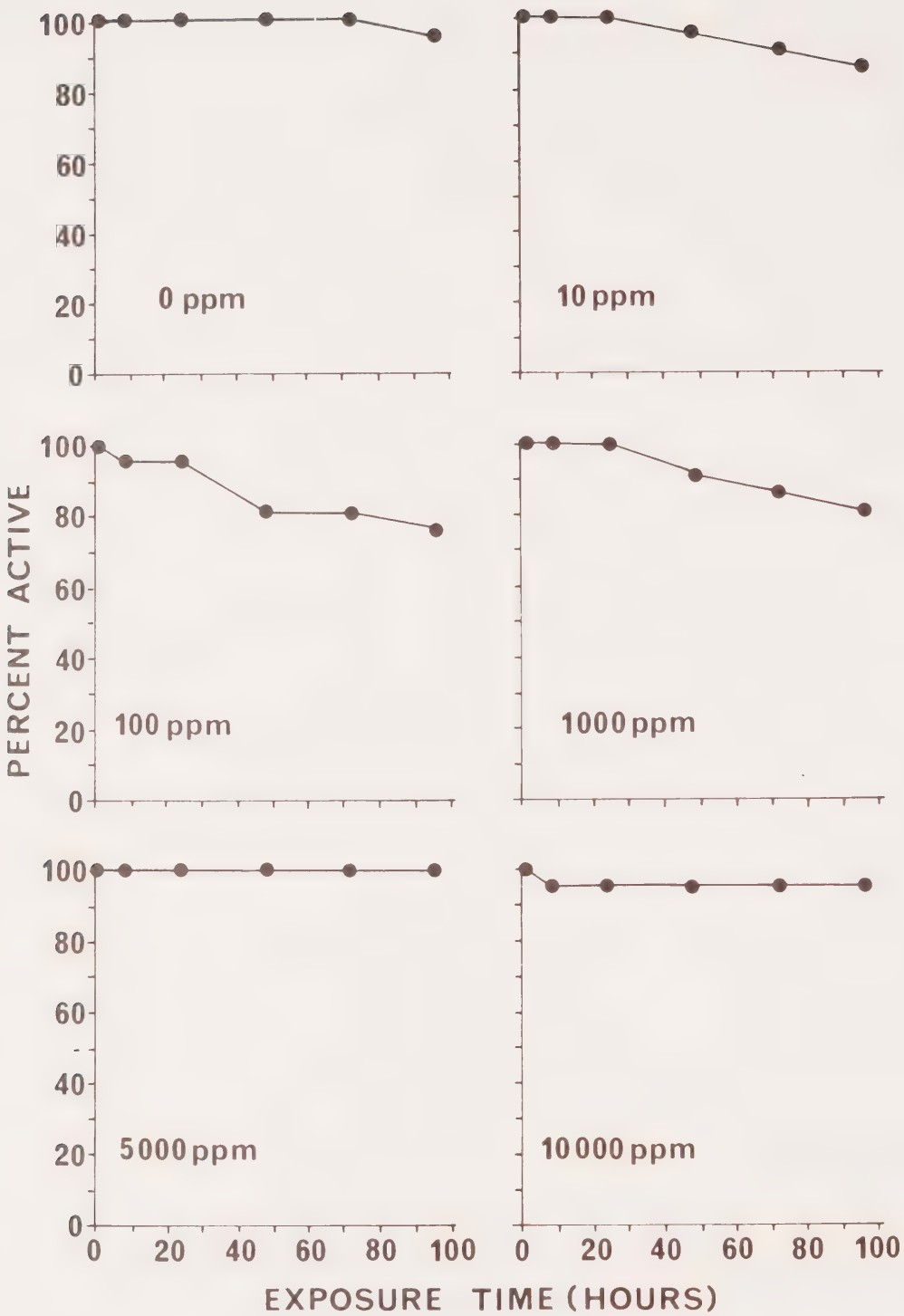

FIGURE 5. Activity of *Brachydiastylis resima* at intervals during 96 hour exposure to different concentrations of Atkinson Point crude oil.

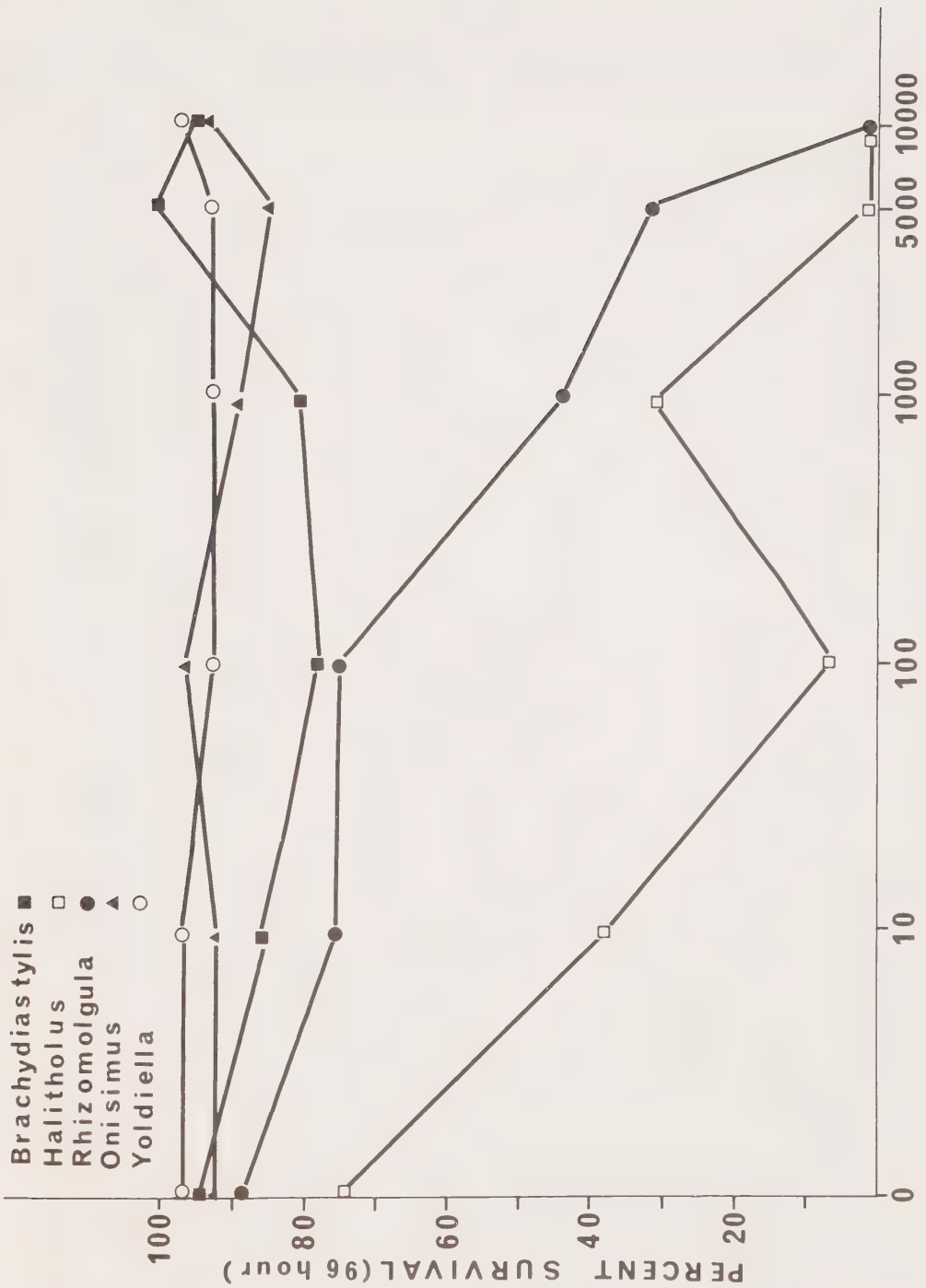

FIGURE 6. Survival of *Onisimus affinis*, *Yoldiella intermedia*, *Rhizomolgula globularis*, *Halitholus cirratus* and *Brachydiastylis resima* after 96 hours exposure to various concentrations of Atkinson Point crude oil.

oil was summarized in Figure 6. It is clear from this figure that Brachydiastylis, Yoldiella and Onisimus are reasonably resistant to even high concentrations of oil in seawater. In contrast Rhizomolgula and Halitholus are rather more susceptible; of the two, Halitholus appears to be the more sensitive and is killed by fairly low concentrations of the oil.

2. Contact Toxicity Tests

This phase of the study examines the ability of the benthic amphipod Onisimus affinis to recover from brief immersion in different types of crude oils followed by a return to clean seawater. Animals in series 1 were exposed to the oil for 30 seconds, while those in series 2 were exposed for two minutes before being rinsed. An activity coefficient defined earlier was used to monitor the general activity of the group at intervals after exposure. In general the activity coefficient in both series 1 (Figure 7) and series 2 (Figure 8) declined progressively following exposure of the animals to the oil. Within hours of transfer of the oiled animals to clean seawater the activity coefficient was reduced by about 5%-30% relative to that of the controls. This short-term effect probably reflects purely physical impairment of locomotion by the viscous oil. From this initial depression the activity level gradually declined until after about four weeks the activity of the group was less than 45% that of controls. An exception is the group exposed to Atkinson Point crude for only 30 seconds. This group exhibits an increase in activity after the first day followed by a reasonably stable level until day 15 when a delayed effect resulted in a rapid decline to a level comparable to that of the groups exposed to Norman Wells and Venezuela crude. In contrast, longer exposure to Atkinson Point crude eliminates this delayed effect and the decline in activity is comparable to that of the other two groups. In general, Norman Wells crude appears to be initially more toxic than Atkinson Point crude, although in the long run the ultimate consequences are similar.

Swimming activity of Onisimus is adversely affected by brief exposure to the oil (Figure 9). Even several days after transfer to clean seawater when visible traces of oil had disappeared from most of the animals only a small percentage of the animals exhibited

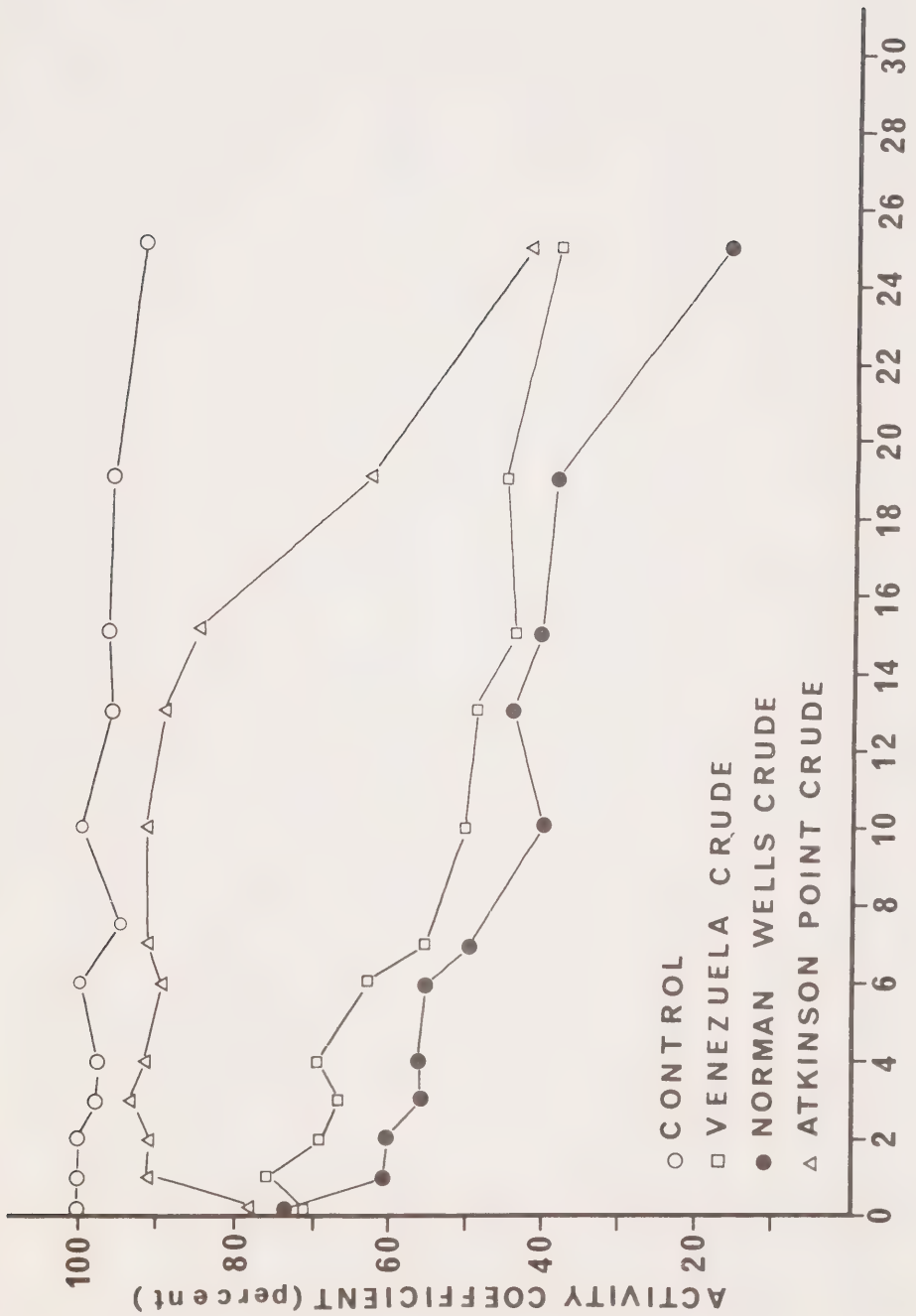

FIGURE 7. Time course of recovery of *Onisimus affinis* following 30 second immersion in Atkinson Point, Norman Wells, and Venezuela crude oils. (Series 1).

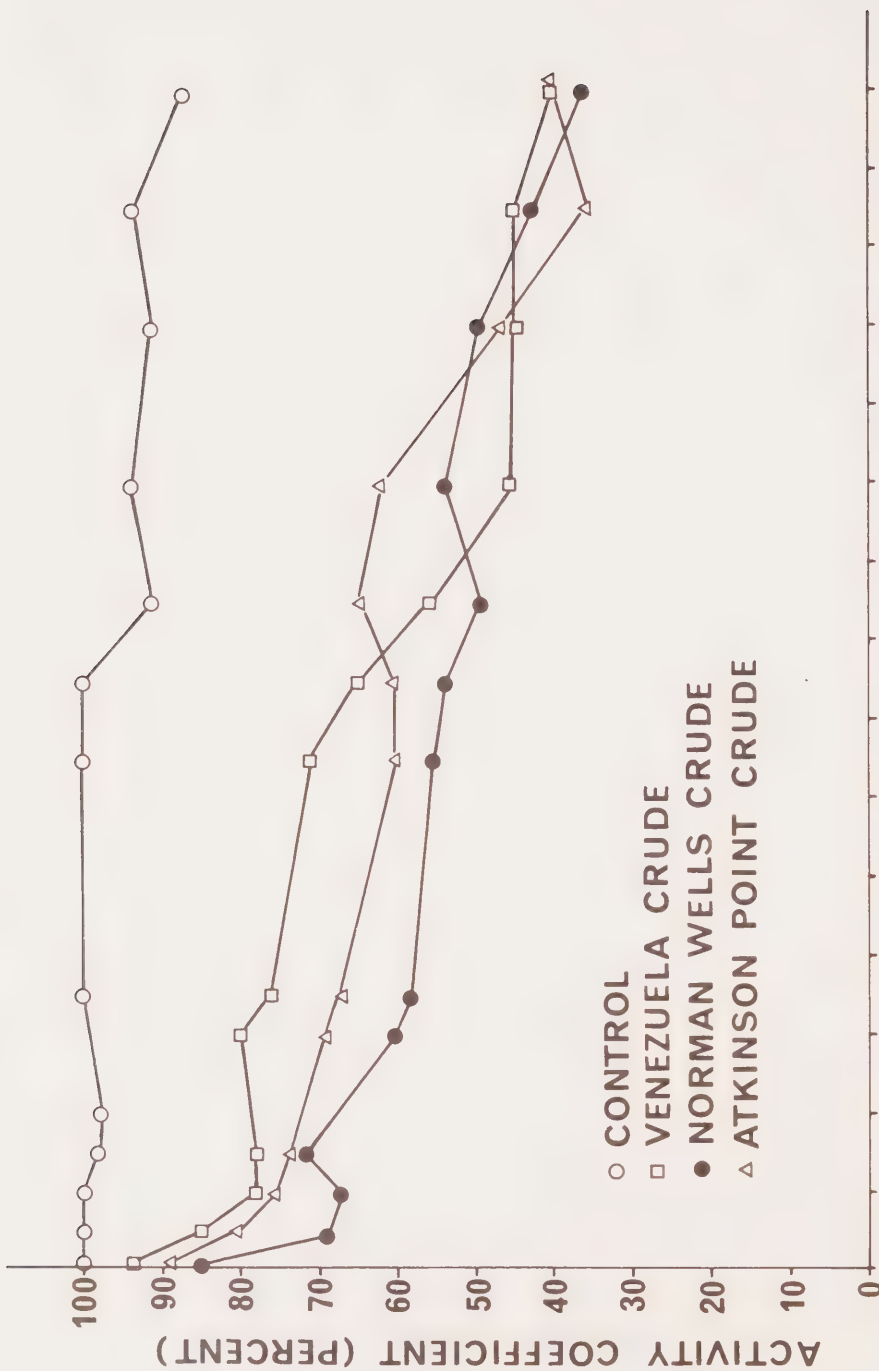

FIGURE 8. Time course of recovery of *Onisimus affinis* following 2 minute immersion in Atkinson Point, Norman Wells, and Venezuela crude oils. (Series 2).

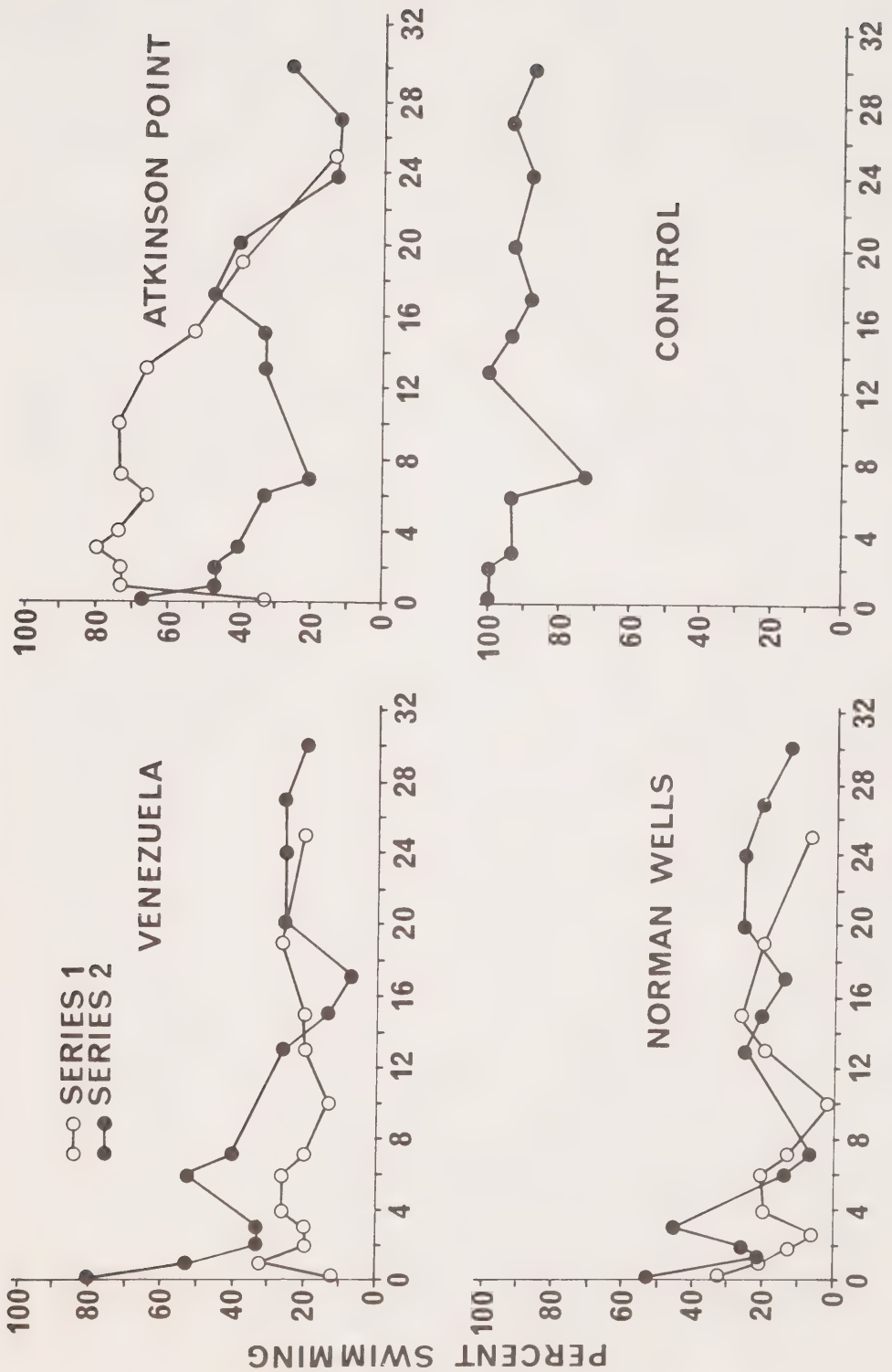

FIGURE 9. Long-term effect of brief immersion in Atkinson Point, Norman Wells, and Venezuela crude oil on the swimming ability of *Onisimus affinis*.

a swimming response. After 25-30 days only about 20% of animals were swimming; and even for these the response was not entirely normal. Many appeared to have difficulty in maintaining their balance. A number of individuals flexed their body in such a manner that the rapid beating of the pleopods caused them to swim on their side in tight circles in brief spasmodic bursts. Very few exhibited the more normal upright swimming posture.

Following transfer to clean seawater a gradual progressive mortality of the oiled animals occurred (Figure 10). Until after about four weeks only 40%-50% of them remained alive.

It is clear that after exposure to crude oil, although individuals are only killed slowly over a long period, their locomatory behavior is severely disrupted from the outset and then progressively deteriorates. Few, if any, of the animals appear capable of recovering fully once exposed to the crude oil.

3. Affinity of Marine Invertebrates for Crude Oils

These experiments examine the behavioral response of several Arctic marine invertebrates to the presence of different types of crude oil in their immediate vicinity. Results for Onisimus affinis, Gammarus oceanicus and Mesidotea entomon are presented in figures 11, 12 and 13, respectively. Each bar in the histogram represents the percentage of the animals observed in the given quarter (indicated by A, B, C and D) of the test chamber during each 30 minute run. The zone containing the oiled sponge is indicated by the dark bar. Each group of four runs constitutes a series for the given animal in the given oil. The dashed line on each graph represents the expected frequency of the animals, assuming random distribution. While it is clear from these graphs that Onisimus and Gammarus generally tend to avoid the oiled zones and that Mesidotea is essentially neutral except with respect to Norman Wells crude, it is difficult to compare the magnitudes of the responses. An affinity coefficient defined earlier provides a useful means of expressing differing degrees of attraction or repulsion. Onisimus is strongly repelled by all three crude oils, with the Arctic oils being considerably more repellant than Venezuela crude. The amphipod Gammarus is also strongly repelled by all three oils, with Atkinson Point oil again being the most repellant.

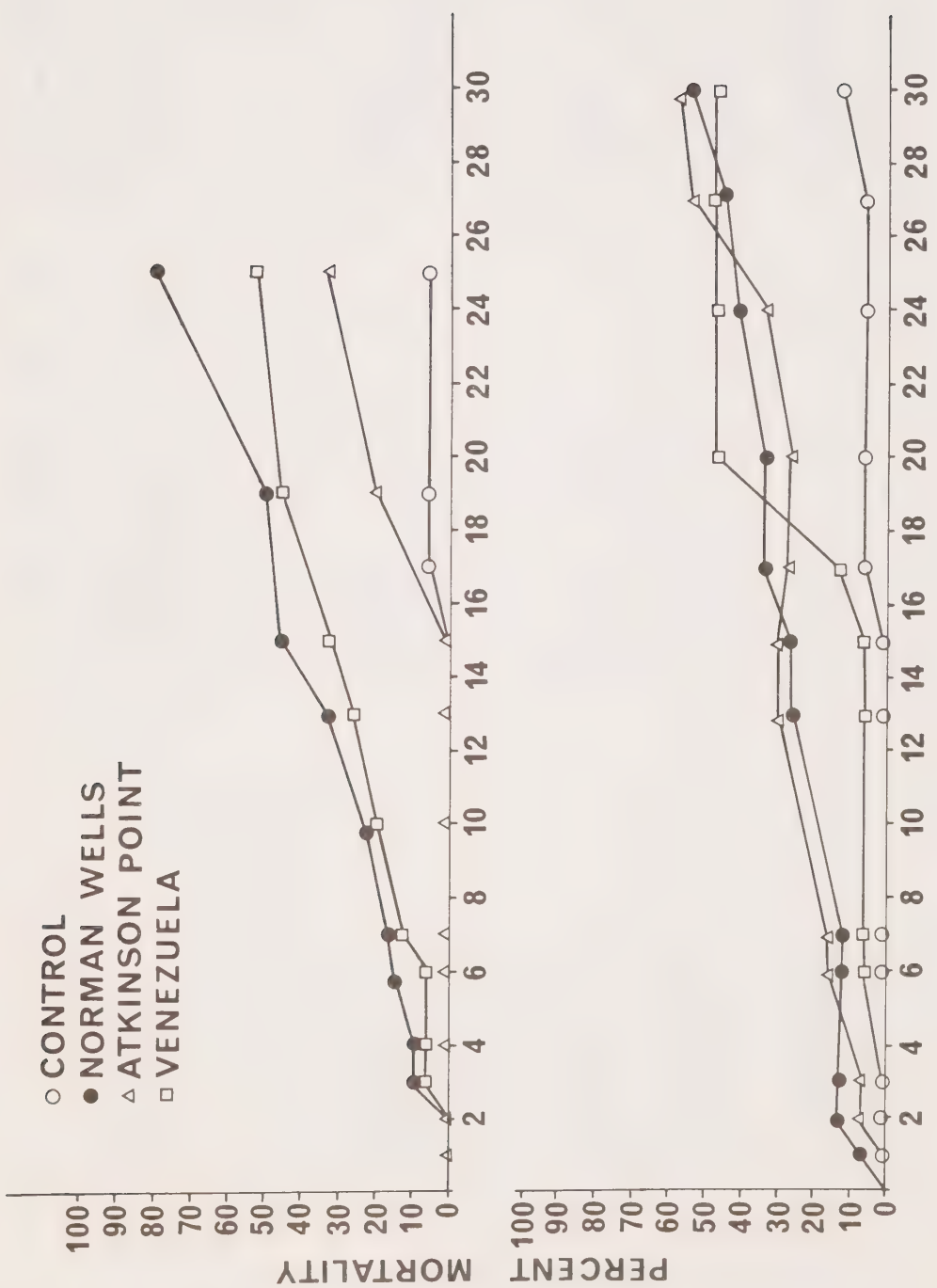

FIGURE 10. Time course of mortality of *Onisimus affinis* following brief immersion in Atkinson Point, Norman Wells, and Venezuela crude oil. (Upper figure series 1; lower figure series 2).

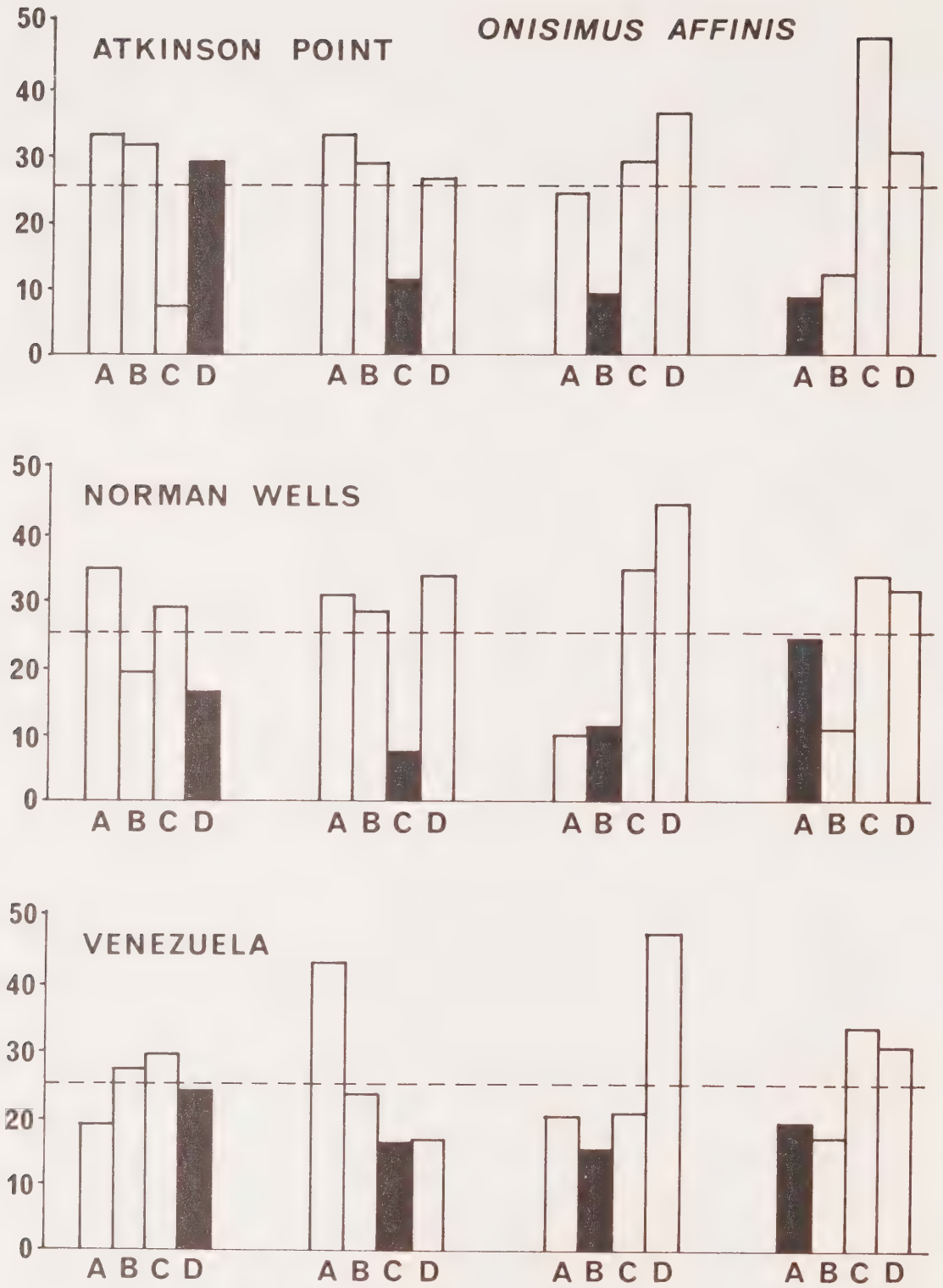

FIGURE 11. Percentage distribution of *Onisimus affinis* in each quarter of the oil affinity test chamber during exposure to various crude oils. Dark bar indicates oiled quarter in each run. Dashed line indicates expected distribution in each quarter.

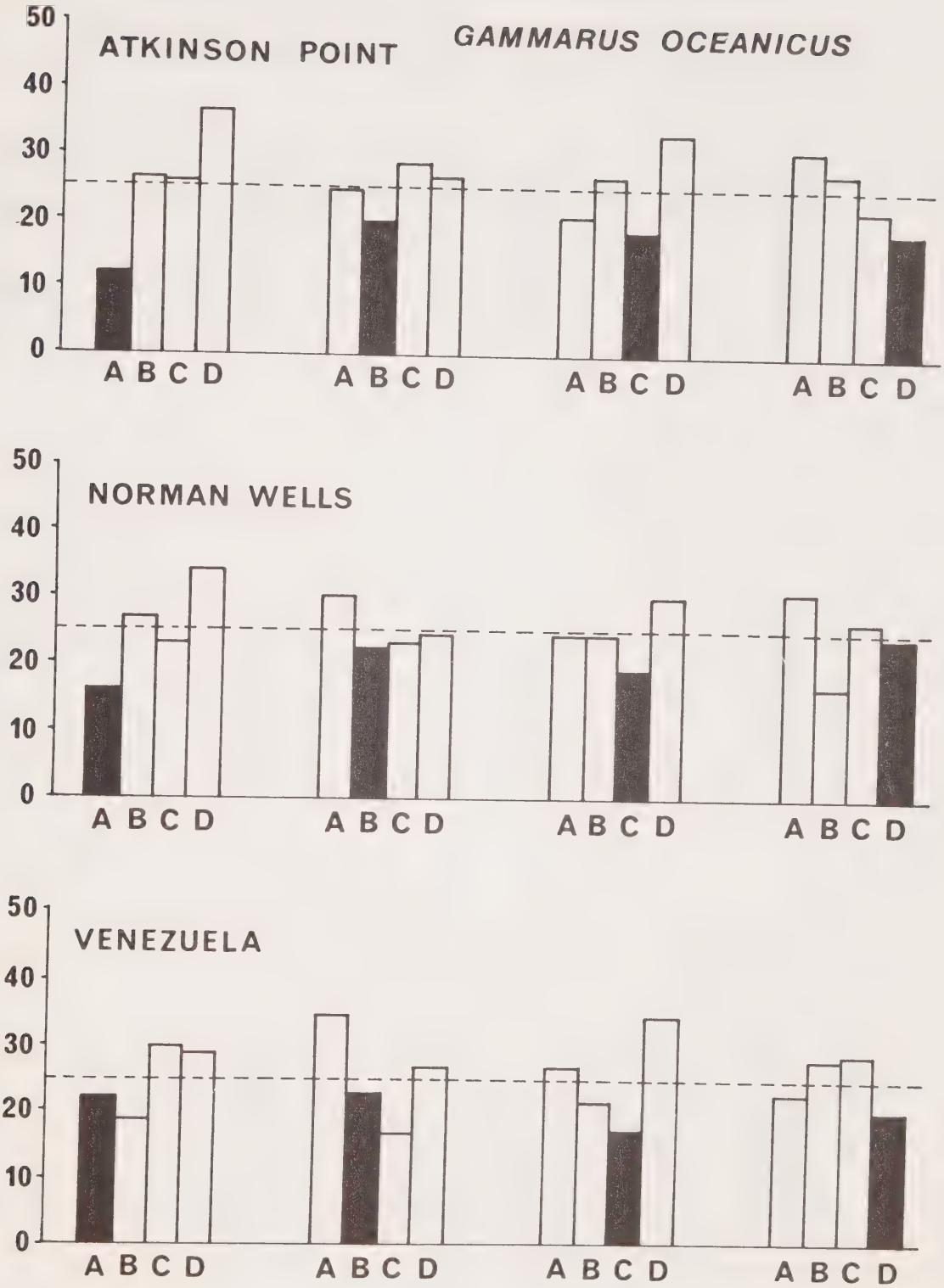

FIGURE 12. Percentage distribution of *Gammarus oceanicus* in each quarter of the oil affinity test chamber during exposure to various crude oils. Dark bar indicates oiled quarter in each run. Dashed line indicates expected distribution in each quarter.

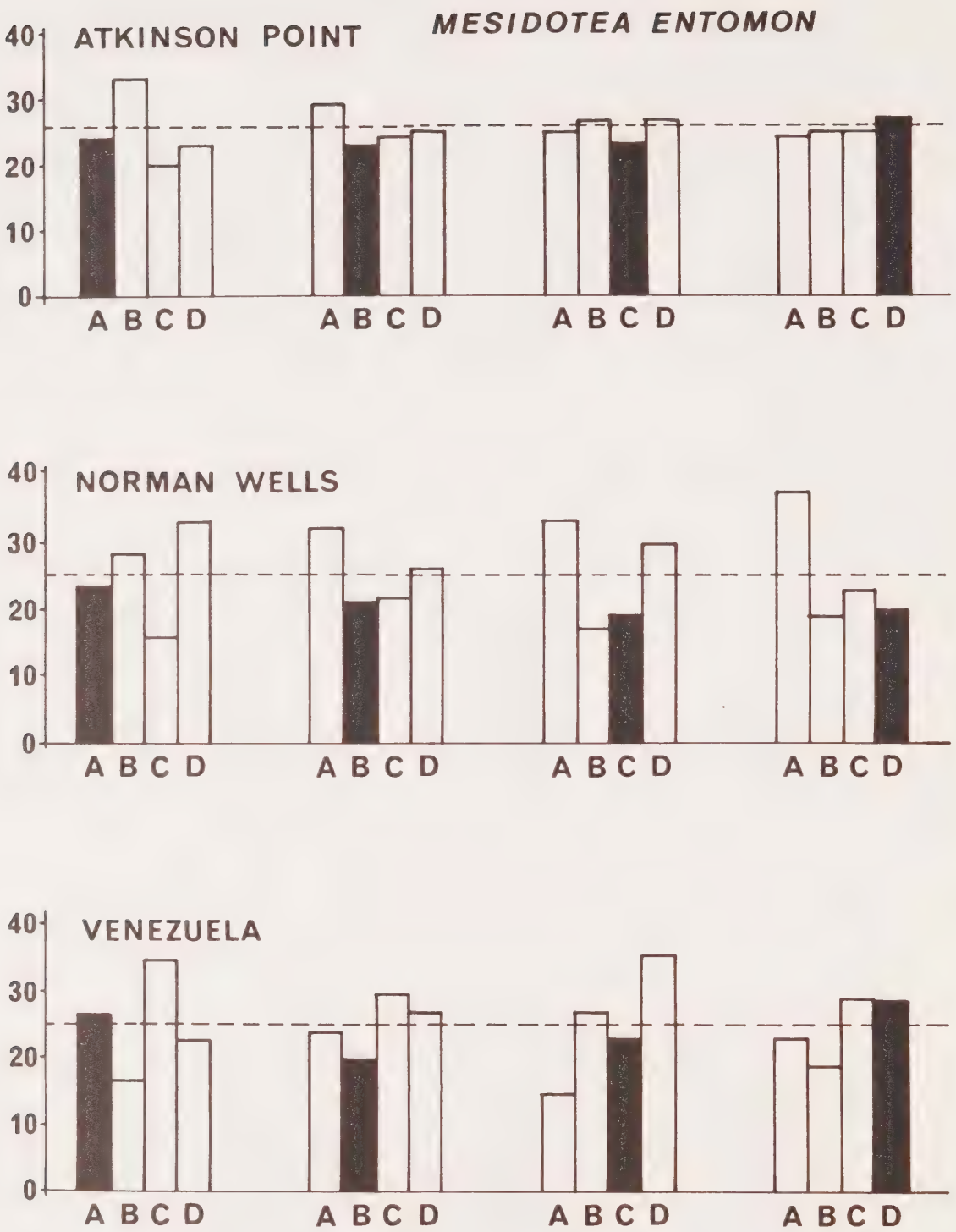

FIGURE 13. Percentage distribution of *Mesidotea entomon* in each quarter of the oil affinity test chamber during exposure to various crude oils. Dark bar indicates oiled quarter in each run. Dashed line indicates expected distribution in each quarter.

TABLE I

Affinity of Onisimus, Gammarus and Mesidotea for Atkinson Point (A.P.), Norman Wells (N.W.) and Venezuela (Ve.) crude oil

SPECIES	OIL TYPE	$\Sigma 0'$	$\Sigma e'$	A.C.	X^2	P
Onisimus	A.P.	176	293	-39.8	103.7	<0.005
Onisimus	N.W.	199	320	-37.9	90.2	<0.005
Onisimus	Ve	228	300	-23.7	30.9	<0.005
Gammarus	A.P.	171	260	-34.2	45.8	<0.005
Gammarus	N.W.	214	260	-17.3	16.3	<0.005
Gammarus	Ve	212	257	-17.4	13.0	<0.05
Mesidotea	A.P.	256	265	-3.3	2.2	N.S.
Mesidotea	N.W.	206	248	-16.9	10.4	<0.05
Mesidotea	Ve	265	271	-2.1	5.7	N.S.

$\Sigma 0'$ = total observed counts in the oiled zone for the given series.

$\Sigma e'$ = total expected counts in the oiled zone for the given series.

A.C. = affinity coefficient expressed as percent.

Norman Wells oil and Venezuela oil are similar in effect. The isopod Mesidotea appears to be essentially neutral to both Atkinson Point oil and Venezuela crude and the affinity coefficients are not significantly different from zero. An A.C. of -16.9 for tests with Norman Wells crude indicates a slight repulsion from the oil, that is probably significant.

None of the animals examined were attracted to oil masses. It is clear that the degree of repulsion varies with both the species and the crude oil type.

4. The Influence of Crude Oil on Metabolic Rate

Data concerning the acute influence of seawater soluble components of Norman Wells and Atkinson Point crude oils upon the metabolic rate of the amphipod Onisimus affinis is presented in Tables II and III. In the presence of low to moderate concentrations of either crude oil the metabolic rate does not differ significantly from that of the control (Figures 14 and 15). At high concentrations both oils stimulate respiration. Atkinson Point oil at concentrations of 1,000 ppm. and 5,000 ppm. increases the metabolic rate by 25% and 30%, respectively. Norman Wells oil at similar concentrations raises the metabolic rate by 25% and 38%, respectively. At still higher concentrations the metabolic rates of animals in both oils decline, that in Atkinson Point only slightly, while that in Norman Wells oil quite significantly.

DISCUSSION

Although the usefulness of short-term toxicity tests as indicators of environmental damage is limited by the fact that they measure only a rather gross physiological change that occurs rapidly, they can, nevertheless, provide valuable information about the relative sensitivity of various species to pollutants. Problems likely to be encountered in conducting such tests have been discussed by Perkins (1972). As pointed out in section 3.4 of the resume of current state of knowledge, species differ markedly in their tolerance of petroleum. Knowledge of such species sensitivity is a prerequisite for predicting general effects of pollution incidents.

TABLE II

The Influence of Various Concentrations of Atkinson Point Crude Oil on the Metabolic Rate of *Onisimus affinis*

OIL CONC. (ppm.)	$\mu l O_2$/ mg/hr.	N	S.D.	S.E.	%ΔC	t	P
0	0.340	10	0.115	0.036	-	-	-
10	0.409	13	0.114	0.032	+20.3	1.37	N.S.
100	0.369	13	0.109	0.030	+8.5	0.59	N.S.
1,000	0.425	16	0.041	0.035	+25.0	1.44	N.S.
5,000	0.443	17	0.146	0.035	+30.3	1.83	<0.05
10,000	0.426	22	0.118	0.025	+25.3	1.82	<0.05

S.D. = Standard deviation

S.E. = Standard error of the mean

%ΔC = % increase over the control

t = Student's statistic

TABLE III

The Influence of Various Concentrations of Norman Wells Crude Oil on the Metabolic Rate of *Onisimus affinis*

OIL CONC. (ppm.)	$\mu l O_2$/ mg/hr.	N	S.D.	S.E.	%ΔC	t	P
0	0.340	10	0.115	0.036	-	-	-
10	0.352	13	0.066	0.018	+3.5	0.30	N.S.
100	0.354	13	0.094	0.026	+4.1	0.30	N.S.
1,000	0.473	14	0.064	0.017	+25.0	3.50	<0.005
5,000	0.469	13	0.035	0.010	+38.0	3.69	<0.005
10,000	0.376	13	0.034	0.009	+10.6	1.03	N.S.

S.D. = Standard deviation

S.E. = Standard error

%ΔC = % increase over the control

t = Student's t statistic

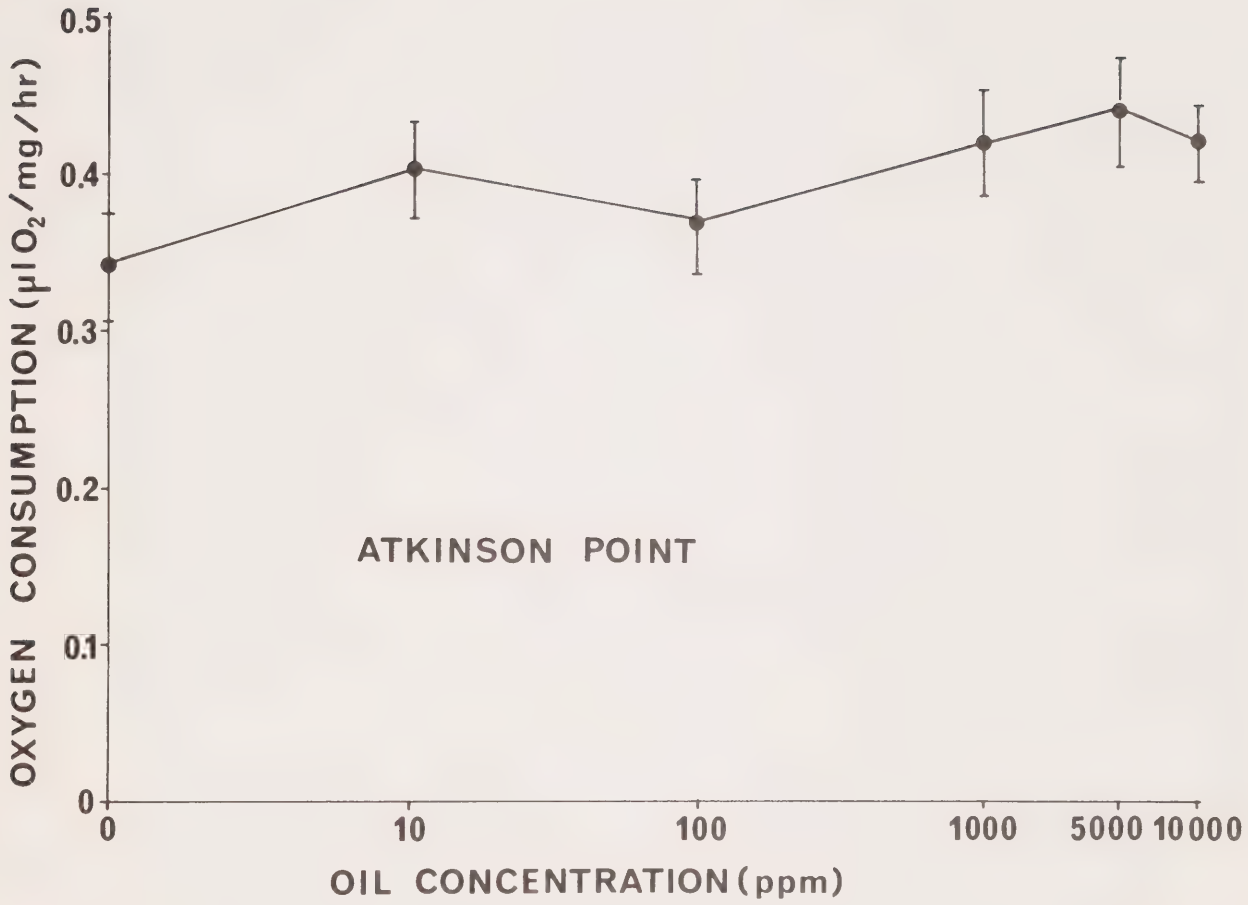

FIGURE 14. Influence of different concentrations of Atkinson Point crude oil on the respiration of *Onisimus affinis*. Vertical lines indicate the standard error of the mean.

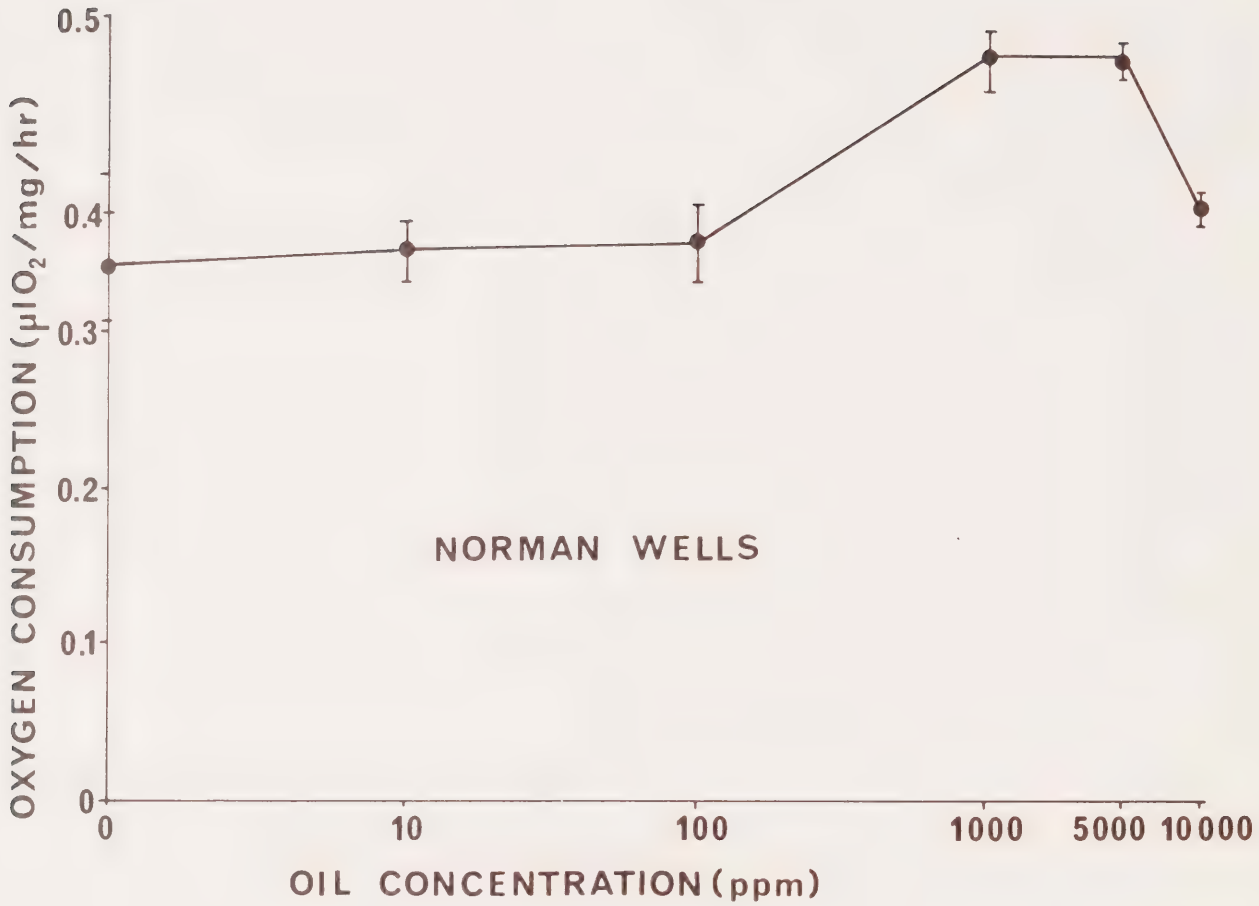

FIGURE 15. Influence of different concentrations of Norman Wells crude oil on the respiration of *Onisimus affinis*. Vertical lines indicate the standard error of the mean.

Particularly sensitive species can sometimes be used as sensitive biological monitors of environmental quality.

Adult forms of the benthic species investigated appear to be rather tolerant of high concentrations of those oil components that dissolve readily in seawater. The concentrations that cause significant mortality are far in excess of those likely to be encountered in the natural habitat, except perhaps in very restricted areas. Moderate concentrations appear to disrupt feeding activity in the mollusc Yoldiella. Interestingly enough, even in the highest concentrations tested the disruption proved to be temporary and the animals resumed activity within four days. Onisimus and Brachdiastylis appear to be very tolerant of petroleum. However, adverse sublethal effects cannot be ruled out. The tunicate Rhizomolgula, a very abundant animal in many areas of the Eskimo Lakes, appears to be more sensitive to crude oil than the other benthic species. However, concentrations that might prove rapidly lethal are unlikely to be attained in the natural habitat. In exposed turbulent areas, emulsification of the oil by wave action and dispersion of the resulting fine droplets in the water column could lead to the ingestion of oil, as Rhizomolgula feeds by filtering particles from the water column. Conover (1971) has shown that such oil droplets can be dispersed to considerable depths and over wide areas, and are readily ingested by filter feeders. The effect of such ingestion of oil upon the animal is not known.

The relatively low tolerance for petroleum exhibited by the planktonic medusa Halitholus must be viewed with some caution in view of the general sensitivity of these animals to capture and confinement. It is possible that a synergistic interaction between experimental stress and petroleum stress rendered them more sensitive to oil than they would actually be in the natural habitat. However, such planktonic forms have generally been found to be particularly sensitive to petroleum. Mironov (1970) found that 100 ppm. crude oil was rapidly lethal to all zooplankton species tested. As discussed in the "Implications" section, Halitholus and other planktonic forms could encounter lethal petroleum in seawater concentrations in the vicinity of an oil spill under ice.

In addition to the toxic effects discussed thus far, oil may adversely affect organisms that come into direct contact with it. The oil by virtue of its high viscosity may physically impair locomotion, feeding

or respiration and thus lead to eventual death. Some animals appear to recover readily from contact with oil. Wilder (1970) noted that lobsters liberally smeared with Bunker C lost most of the oil within a matter of hours and suffered no subsequent mortality over a 35 day period. In this instance it must be kept in mind that Bunker C lacks the lighter, more toxic fractions that are present in crude oils. Onisimus does not readily recover from oiling and a high percentage of the animals eventually succumb, even though in most instances visible traces of the oil disappear in a few days. Activity of the animal is severely disrupted by the treatment. Few of the animals are capable of swimming, and those that are appear to have difficulty maintaining balance. Mortality occurs gradually over an extended period. Indications are that the adverse effects result from the presence of relatively insoluble toxic components of the oil rather than from a smothering effect. On the basis of these results, contact with sub-ice oil masses probably poses the greatest potential threat to Onisimus as discussed in the section on "Implications".

In view of the differential sensitivity of various species to crude oil it is clear that an Arctic oil spill of some magnitude would probably selectively eliminate certain species from an area. We know too little about the ecological inter-relationships of Arctic organisms to even hazard a guess as to the broader ramifications of such a disturbance of the ecological balance.

Few studies have investigated the behavioral response of marine invertebrates to the presence of crude oil. As pointed out elsewhere (No. 6 of "Resume of Current State of Knowledge") field observations have suggested that some species actively avoid oil while others may in fact be attracted to it. In view of the high mortality of Onisimus following even very brief contact with the oil, the behavioral response of the animal in the presence of oil is of interest, particularly as many of the animals congregate in areas where subice pockets of oil could be expected to accumulate. Both of the amphipod species tested were clearly repelled by the oil, the degreee of repulsion varying somewhat with the type of oil. It is possible that after extended exposure to sublethal concentrations of the oil the animals avoidance reaction may diminish either through adaptation or damage to chemoreceptors. We are presently investigating this possibility. Yoldiella was found to exhibit an initial avoidance response (shell closure)

followed by a gradual return to normal activity in the continued presence of oil. Mesidotea shows little if any response to the oil. None of the species examined exhibited the slightest degree of attraction for any of the crude oils. Little is known regarding other possible behavioral effects of dissolved crude oil.

Even those species that are classed as tolerant of crude oil on the basis of short-term toxicity tests may still be adversely affected and eventually succumb over a more extended exposure period. For this reason, more sensitive physiological criteria are required for detecting sublethal deleterious effects. Metabolic rate is a sensitive indicator of an organisms general physiological state. Neither Atkinson Point nor Norman Wells crude had any significant acute effect on respiration of the amphipod Onisimus at low or moderate concentrations. At concentrations of 1,000 ppm. and 5,000 ppm. Atkinson Point crude increased respiration by 25 and 30 per cent, respectively. At similar concentrations Norman Wells oil increases respiration by 25% and 38%, respectively. At still higher concentrations the rates decline somewhat in both oils. The increased respiration may reflect an increase in the animals' activity rather than a direct effect on metabolic processes. Both the metabolic rate and the rate of crawling of the intertidal snail Littorina littorea increased significantly upon exposure to high concentrations of Bunker C (Hargrave and Newcombe, 1973). It is possible that in both Onisimus and Littorina an escape response is involved. If such is the case it is likely that on longer exposure to the pollutant the metabolic rate would decrease. Petroleum certainly does not appear to have a depressant effect upon the activity of Onisimus, as has been reported for other invertebrates (Galtsoff, 1935; Smith, 1970). A significant decline in activity would have been accompanied by a depression of respiration.

CONCLUSIONS

Arctic marine invertebrates vary considerably in their tolerance of crude oils. Benthic species are generally tolerant of high concentrations of seawater soluble components of the oil. Indications are that planktonic species may be more sensitive and are killed at low to moderate concentrations of the oil.

Certain benthic species that are tolerant of seawater-soluble components of the oil are susceptible to physical contact with the oil. Little recovery occurs after transfer to clean seawater. Activity is severely depressed and mortality occurs gradually.

A number of benthic species actively avoid crude oil. The degree of avoidance varies with both the species and with the type of crude oil. Arctic oils generally appear to be more repellant than Venezuela crude. The avoidance response may be of particular ecological significance to species living in the sub-ice habitat.

Acute sublethal effects of the metabolism of Onisimus are not detectable at petroleum concentrations likely to be encountered in the natural habitat. An increased rate at very high petroleum levels probably reflects an increase in activity associated with an escape response.

In view of the absence of well developed littoral communities in the Arctic it is concluded that the major immediate impact of oil spills is likely to be upon the planktonic and epibenthic species that constitute the sub-ice community. The precise ecological consequence of extensive damage to the sub-ice community is at present unknown.

The overall general effect of a major oil spill will probably be the selective elimination of sensitive species from the habitat coupled with an increase in numbers of the more tolerant species as competition decreases. The natural balance of the ecosystem will probably not be re-established for several years.

IMPLICATIONS: CRUDE OIL AND MARINE BIOLOGY IN THE ARCTIC CONTEXT

In the Arctic Ocean, ice abrasion effectively inhibits the establishment of littoral communities in all but the most protected areas. Thus the smothering and contact poisoning by amorphous masses of beached oil that play such a significant destructive role in temperate zone pollution incidents will have little direct impact upon marine invertebrate populations of the Arctic.

Sublittoral areas will undoubtedly be of greater significance from the point of view of potential biological damage.

As was pointed out earlier, in the absence of far northern refining facilities it is the persistent petroleum pollutants (crude oils) that represent the greatest potential threat to the Arctic marine ecosystem. It is generally agreed* that crude oils and heavy residual products are on the whole less immediately damaging to marine ecosystems (excluding perhaps intertidal habitats) than many of the lighter refined products. This certainly appears to be the conclusion to be drawn from a number of major spills in temperate waters. Thus, the Torrey Canyon spill (Smith, 1970), the Santa Barbara spill (Foster et al., 1971) and the Arrow spill (Scarratt, 1970; Thomas, 1970) involving Kuwait crude, California crude and Bunker C oil, respectively, appear to have resulted in relatively minor destruction of sublittoral communities. In contrast, the Buzzards Bay spill (Blumer et al., 1970) and the Tampico Maru spill (North et al., 1964) involving No. 2 fuel oil and diesel oil, respectively, resulted in wholesale destruction of littoral and sublittoral communities.

In view of the above considerations it might be concluded that crude oil spills would have little significant effect on Arctic marine invertebrate communities. However, a number of unique features of polar ecosystems may render them acutely susceptible to serious damage by spilled oil.

The sub-ice communities may be particularly vulnerable to oil pollution damage. In the spring, a dense algal bloom generally forms on and within the undersurface of the ice, stimulated both by the increasing levels of nutrients in the water column at this time of year and by the increasing levels of diffuse light filtering through the ice. As the bloom progresses the bottom of the ice may become quite brown. There is evidence that large numbers of invertebrate animals congregate in the vicinity of the lower ice surface, presumably for the purpose of feeding on the algal layer. This sub-ice community, which needs to be studied

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*A number of exceptions have been noted in No. 1 of "Resume of Current State of Knowledge".

in much greater detail, includes planktonic species as might be expected. Surprisingly, however, we have also found large numbers of individuals of epibenthic species that normally are found in close proximity to the sea floor. For example, in the Eskimo Lakes, we found the Onisimus affinis population divided into a large sub-ice group and a slightly larger benthic group, with few, if any, individuals occurring at intermediate layers in the water column. Crude oil, being heavier than ice yet lighter than seawater tends to spread under the ice and collect in pockets at the interface (Hoult, 1972). There are several potential ecological effects that need to be examined carefully. Oil has been shown to be toxic to phytoplankton (Mironov, 1972) and thus could prevent or inhibit formation of the sub-ice flora, with unknown consequences both to the animals dependent upon it and to the later diatom bloom in the water column. Furthermore, the oil could physically isolate herbivorous animals from their algal food, again with unknown long-term consequences. Finally, the animals would be particularly exposed to the hazards of contact poisoning and to possible toxic effects arising from the ingestion of quantities of crude oil. We have demonstrated that Onisimus affinis exhibits a very low degree of recovery following even very brief contact with crude oil. However, we have also found that this species tends to avoid crude oil masses. It would be useful to obtain similar data for other members of the sub-ice community and to carry out field observations to confirm that such an avoidance response is operative in the natural sub-ice habitat.

In temperate and tropical waters spilled crude oil appears to lose much of its toxicity fairly rapidly by evapoation of the light more toxic fractions. Thus, the Kuwait crude oil spilled from the Torrey Canyon was estimated to have lost about 25% of its volume through evaporation during the first few days (Smith, 1970). Such rapid weathering probably accounts for the observations already alluded to that crude oil spills as a rule do not cause disastrous damage to subtidal communities. Presumably, the rapid loss of volatile material results in a considerable reduction in the amounts of the lighter, toxic fractions available for dissolution in the seawater. A spill in Arctic waters might have far different results. The lower temperature would reduce the rate of evaporation of toxic fractions. If the spill occurs in an under-ice situation the physical barrier of the ice would probably further minimize evaporation. The oil would retain its

toxicity for an extended period and it is probable that higher concentrations of the soluble toxic components would ultimately dissolve in the seawater. The more sensitive species of the plankton and of the sub-ice community might thus be exposed to lethal levels of the pollutant. As we have shown, Halitholus a common planktonic medusa suffers high mortality at concentrations of 100 ppm. Arctic crude oil. Similarly, Mironov (1970) found that all of the zooplankton species tested (mostly copepods) suffered high mortality within 24 hours at concentrations of 100 ppm crude oil. Larval stages of many species also occur as members of the plankton community. It has been shown that many marine invertebrate larvae are far more sensitive to petroleum than are adults, often succumbing at concentrations in the range 1-100 ppm. (Wells, 1972; Mironov, 1970).

As McLaren (1964) and others have pointed out, it is primarily the very abbreviated season of phytoplankton abundance rather than the frigid temperature that makes the Arctic Ocean a particularly harsh environment. Many Arctic marine invertebrates feed intensively during this brief surge of phytoplankton productivity. At this time, many appear to accumulate nutrient reserves, in the form of lipids or glycogen, to sustain them through the ensuing winter. Furthermore, some species are probably dependent upon the accumulated reserves for the production of nutrient rich eggs that are released during the winter or early spring. Many studies have shown that in certain species oil, while not causing significant immediate mortality may severely inhibit the accumulation of nutrient reserves by disrupting feeding. Galtsoff et al. (1935) found that oil has an anaesthetic effect on the gill cilia of oysters. The net result was a reduction in pumping, a decrease in feeding and a consequent cessation of growth and glycogen deposition. The present study suggests that moderate concentrations of oil disrupt the feeding behaviour of Rhizomolgula and Yoldiella; although in the latter case recovery of activity appears to occur rapidly. To properly assess the ecological significance of such nutritional effects we require considerably more information about the influence of sublethal concentrations of oil on a wider range of invertebrate species. In the Arctic, such a disruption of feeding by oil for even a relatively brief period during the time of maximum phytoplankton availability could seriously impair the ability of a population to survive the winter or to produce viable eggs in the spring.

Perhaps the most disturbing feature concerning Arctic oil pollution is the fact that once a given marine community is severely damaged prospects for recovery within a reasonable time are slight. As Chia (1970) has pointed out, a major pollution incident not only eliminates adults but also the existing stock of more sensitive larval stages. Toxic products could be expected to persist in the habitat for a considerable period. Replacement of species from adjacent areas would probably be slow and "with the slow growth and development of sexual maturity of these animals, the re-establishment of the community with a balanced age structure will require many more years than in communities in temperate waters." (Chia, 1970).

LITERATURE CITED

Allen, H. 1971.
Effects of petroleum fractions on the early development of a sea urchin. Mar. Poll. Bull. 2(9): 138-140.

Alyakrinskaya, I.O. 1966.
On the behavior and ability to filter of the Black Sea mussel Mytilus galloprovincialis in oil polluted water. Zool. Zh. 45:998-1003.

Blumer, M. 1970.
Oil contamination and the living resources of the sea. In: FAO Technical Conference on Marine Pollution and its Effects on Living Resources and Fishing. Food and Agriculture Organisation of the United Nations. Dec. 9-18, 1970, Rome.

Blumer, M., H.L. Sanders, J.F. Grassle, and G.R. Hampson. 1971.
A small oil spill. Environment 13(2):2-;3.

Butler, M.J.A. and F. Berkes. 1972.
Biological aspects of oil pollution in the marine environment. A Review. Marine Sciences Centre, McGill University, Montreal. Manuscript Report No. 22.

Chia, F.S. 1970.
Reproduction of Arctic marine invertebrates. Mar. Poll. Bull., 1 (NS) (5): 78-79.

Chia, F.S. 1973.
Killing of marine larvae by diesel oil. Mar. Poll. Bull. 4(2): 29-30.

Chipman, W.A., and P.S. Galtsoff. 1949.
Effects of oil mixed with carbonized sand on aquatic animals. Spec. Scient. Rep. U.S. Fish. Wildlife Serv., 1:1-53.

Conover, R.J. 1971.
Some relations between zooplankton and Bunker C oil in Chedabucto Bay following the wreck of the

tanker "Arrow". J. Fish. Res. Bd. Canada 28:1327-1330.

Crapp, G.B. 1969.
Second report by zoologist. Annual Report of Oil Pollution Research Unit, 1968: Z1-Z24. Field Studies Council, Orielton.

Crapp, G.B. 1971.
Field experiments with oil and emulsifiers. In: The Ecological Effects of Oil Pollution on Littoral Communities (E.B. Cowell, ed.). The Institute of Petroleum, London/ pp. 114-128.

Dean, R.A. 1968.
The chemistry of crude oils in relation to their spillage on the sea. Fld. Stud., 2 (suppl): 1-6.

Dunbar, M.J. 1968.
Ecological development in polar regions: A study in evolution. Prentice-Hall, New Jersey, 113 pp.

Dunbar, M.J. 1971.
Environment and good sense. McGill-Queens University Press, Montreal, 92 pp.

Foster, M.' M. Neushul, E.R. Zingmark. 1971
The Santa Barbara oil spill. Part 2: Initial effects on intertidal and kelp bed organisms. Environ. Poll. 2: 115-134.

Galtsoff, P.S., H.F. Prytherch, R.O. Smith, S.V. Koehring. 1935.
Effects of crude oil pollution on oysters in Louisiana waters. Bull. Bur. Fish. Wash. 18:143-210.

Galtsoff, P.S. 1936.
Oil pollution in coastal waters. Proc. N. Amer. Wildlife Conf. 1:550-55

Goltsoff, P.S. 1964.
The American Oyster, Crassostrea virginica Gmelin. Bur. Comm. Fish., U.S. Fish and Wildlife Serv., Fishery Bull. 64.

Hargrave, B.T. and C.O. Newcombe 1973.
Crawling and respiration as indices of sublethal effects of oil and a dispersant on an intertidal snail, Littorina littorea. J. Fish. Res. Bd. Canada 31(12):1789-1792.

Hawkes, A.L. 1961.
A review of the nature and extent of damage caused by oil pollution at sea. Trans. N. Amer. Wildlife Nat. Resources Conf., 26:343-355.

Hodgmen, C.D., R.C. Weast and S.M. Selby (Editors). 1960.
Handbook of Chemistry and Physics. Chemical Rubber Co., Cleveland.

Hoult, D.P. 1972.
Marine pollution, concentrating on the effects of hydrocarbons in seawater. Canadian-U.S. Maritime Problems and Policies and the Implication for the Development of International Law. Workshop Papers. (L.M. Alexander and G.R.S. Hawkins, eds.) Univ. Rhode Island, Law of the Sea Institute. Kingston pp. 29-31.

Kuhnhold, W.W. 1970.
The influence of crude oils on fish fry. In: FAO Technical Conference on Marine Pollution and its Effects on Living Resources and Fishing. Food and Agriculture. Organization of the United Nations, Dec. 9-18, 1970, Rome.

Lewis, J.B. 1971.
Effect of crude oil and oil spill dispersants on reef corals. Mar. Poll. Bull. 2(4):59-62.

Manwell, C., and C.M. Baker. 1967.
Oil and detergent pollution. The Journal of the

Devon Trust for Nature Conservation. (Supplement): 39-72.

McKee, J.E. 1956.
Oily substances and their effects on the beneficial uses of water. Publ. No. 16, Calif. State Water Pollution Control Bd., Sacramento.

McLaren, I.A. 1964.
Marine life in Arctic waters. In: The Unbelievable Land (I.N.S. Smith, Ed.). Queen's Printer, Ottawa, pp. 93-97.

Mironov, O.G. 1967.
The effect of oil and oil products upon some molluscs in the Littoral zone of the Black Sea. Zool. Zh., 46:134-136.

Mironov, O.G. 1969.
The effect of oil pollution upon some representatives of the Black Sea zooplankton. Zool. Zh. 48:980-984.

Mironov, O.G. 1970.
The effect of oil pollution on flora and fauna of the Black Sea. In: FAO Technical Conference on Marine Pollution and its Effects on Living Resources and Fishing. Food and Agriculture Organization of the United Nations. Dec. 9-18, 1970, Rome.

Naylor, E., 1965.
Biological Effects on a Heated Effluent in Docks at Swansea, South Wales. Proc. Zool. Soc. London 144:253-68.

Nelson-Smith, A. 1968.
The effects of oil pollution and emulsifiers cleansing on shore life in Southwest Britain. J. Appl. Ecol. 5:97-107.

Nelson-Smith, A. 1970.
The problem of oil pollution of the sea. Adv. Mar. Bio., 8:215-306.

Nelson-Smith, A. 1973.
Effects of oil on marine plants and animals. In: Water Pollution by Oil. (P. Hepple, editor) The Institute of Petroleum, London pp. 273-280.

North, W.J., M. Neushul and K.A. Clendenning. 1964.
Successive biological changes observed in a marine cove exposed to a large spillage of mineral oil. Symp. Poll. Mar. Micro-org. Prod. Petrol. Monaco, pp. 335-354.

Ottway, S. 1971. The comparative toxicities of crude oils. In: The Ecological Effects of Oil Pollution on Littoral Communities. (E.B. Cowell, Ed.) The Institute of Petroleum, London, pp. 172-180.

Perkins, E.J. 1972.
Some problems of marine toxicity studies. Mar. Poll. Bull. 3(1):13-14.

Scarratt, D.J. 1970.
Sublittoral biological survey team summary report May 13, 1970. Unpublished report. Fisheries Research Board, St. Andrews, New Brunswick.

Shelton, R.G. 1971.
Effects of oil and oil dispersant on the marine environment. Proc. R. Soc. (B), 177:411-422.

Smith, J.E. (ed). 1970.
"Torrey Canyon" Pollution and Marine Life: A Report by the Plymouth Laboratory of the Marine Biological Association of the United Kingdom, Cambridge University Press, 196 pp.

Spiegel, MR. 1961.
Theory and Problems of Statistics. Schaum Publ., New York.

Tegelberg, H. 1964.
Washington's razor-clam fisheries in 1964. Rep. Wash. State Dept. Fisheries, 74:53-56.

Thomas, M.L.H. 1970.
Effects of Bunker C oil on intertidal and lagoonal organisms in Chedabucto Bay, Nova Scotia. Unpublished Report. Marine Ecology Laboratory, Dartmouth, Nova Scotia.

Van Overbeek, J. and R. Blondeau. 1954.
Mode of action of phytotoxic oils. Weeds, 3:55-65.

Vernberg, W.B. and F.J. Vernberg,. 1972.
The synergistic effects of temperature, salinity and mercury on survival and metabolism of the adult fiddler crab, Uca pugilator. Fish. Bull. U.S. 70:415-420.

Wardley-Smith, J. 1968.
Problems in dealing with oil pollution on sea and land. In: Scientific Aspects of Pollution of the Sea by Oil. P. Hepple (ed.) Institute of Petroleum, London, pp. 60-67.

Wells, P.G. 1972.
Influence of Venezuelan crude oil on lobster larvae. Mar. Poll. Bull. 3(7):105-106.

Wilder, D.G. 1970.
The tainting of lobster meat by Bunker C oil alone or in combination with the dispersant Corexit. Unpublished Report. Fisheries Research Board of Canada, St. Andrews, New Brunswick.

Zitko, V. and W.V. Carson. 1970.
Project oil chemistry reports. Unpublished Report. Fisheries Research Board of Canada, St. Andrews, New Brunswick.

TABLE IV

Observed and Expected Distributions of
Gammarus oceanicus in Oiled and
Unoiled Zones During Each Run

OIL TYPE	OILED ZONE	COUNTS 0'	COUNTS 0_c	TOTAL OBSERVED COUNTS	COUNTS e'	COUNTS e_c	X^2
A.P.	a	30	226	256	64	192	24.08
A.P.	b	45	179	224	56	168	2.88
A.P.	c	48	208	256	64	192	5.33
A.P.	d	48	255	303	76	228	13.51
N.W.	a	44	228	272	68	204	11.30
N.W.	b	57	198	255	63.8	191.2	0.97
N.W.	c	50	205	255	63.8	191.2	3.98
N.W.	d	63	193	256	64	192	0.02
Ve	a	57	199	256	64	192	1.02
Ve	b	58	198	256	64	192	0.75
Ve	c	44	212	256	64	192	8.33
Ve	d	53	206	259	64.8	194.2	2.87

COUNTS 0' = observed counts in oiled zone.

COUNTS 0_c = observed counts in control zones.

COUNTS e' = expected counts in oiled zone.

COUNTS e_c = expected counts in control zones.

TABLE V

Observed and Expected Distributions of *Onisimus affinis* in oiled and unoiled zones during each run

OIL TYPE	OILED ZONE	COUNTS 0'	COUNTS 0_c	TOTAL COUNTS	COUNTS e'	COUNTS e_c	X^2
A.P.	a	23	233	256	64	192	35.02
A.P.	b	31	284	315	78.8	236.2	38.67
A.P.	c	34	258	292	73	219	27.78
A.P.	d	88	219	307	76.8	230.2	2.18
N.W.	a	87	265	352	88	264	0.02
N.W.	b	34	274	308	77	231	32.02
N.W.	c	20	252	272	68	204	11.29
N.W.	d	58	291	349	87.2	261.8	13.04
Ve	a	58	244	302	75.5	226.5	5.41
Ve	b	47	257	304	76	228	14.76
Ve	c	48	240	288	72	216	10.67
Ve	d	75	229	304	76	228	0.02

COUNTS 0' = observed counts in oiled zone.

COUNTS 0_c = observed counts in control zones.

COUNTS e' = expected counts in oiled zone.

COUNTS e_c = expected counts in control zones.

TABLE VI

Observed and Expected Distributions of
Mesidotea entomon in oiled and
unoiled zones during each run

OIL TYPE	OILED ZONE	COUNTS 0'	COUNTS 0_c	TOTAL OBSERVED COUNTS	COUNTS e'	COUNTS e_c	X^2
A.P.	a	64	199	263	64.8	198.2	0.01
A.P.	b	61	211	272	68	204	0.96
A.P.	c	58	197	255	63.8	191.2	0.70
A.P.	d	73	199	272	68	204	0.49
N.W.	a	55	184	239	59.8	179.2	0.51
N.W.	b	50	190	240	60	180	2.22
N.W.	c	49	207	256	64	192	4.69
N.W.	d	52	204	256	64	192	3.00
Ve	a	72	200	272	68	204	0.16
Ve	b	54	218	272	68	204	3.84
Ve	c	61	206	267	66.8	200.2	0.67
Ve	d	78	193	271	67.8	203.2	1.02

COUNTS 0' = observed counts in oiled zone.

COUNTS 0_c = observed counts in control zones.

COUNTS e' = expected counts in oiled zone.

COUNTS e_c = expected counts in control zone.

Part IIIa

Baseline Study of the Marine Environment in the Area of the Mackenzie Delta

by

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February 1974

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

SUMMARY

A 12-day cruise on the "North Star of Herschel Island" was carried out in the south Beaufort Sea in late July of 1973. More than half the planned sampling stations were not reached because the sea-ice remained within about 25 miles of shore along much of the coast. Consequently only inshore waters were sampled.

Temperature and salinity distribution in the south Beaufort Sea showed conditions typical of waters off the mouths of large rivers. Relatively warm water of low salinity was found to extend from the river mouths over underlying colder, more saline water. Phosphate-phosphorus, nitrate-nitrogen and silicate-silicon, found to be most abundant in surface waters nearest river mouths, were obviously contributed to the Beaufort Sea from the Mackenzie River. Oxygen and chlorophyll data indicated a low inshore primary production rate. It is concluded that lack of light for photosynthesis, brought about by high river-contributed turbidity, was probably the major limit on production in the nutrient-rich inshore waters.

At least 45 zooplankton species were found in the south Beaufort Sea. Their number suggests a greater faunal diversity than most of the region in fact supports. At 75% of the stations there were only 13 species found in all. However, at the two stations with the greatest diversity there were 36. The relatively large number of low-diversity stations appear also to have supported a low biomass. They were in the region of maximal Mackenzie River influence during the last week of July, 1973, and the few species (crustaceans Cyclops, Limnocalanus, Eurytemora, Mysis, and a few others) are characteristic of fresh to moderately brackish water. A few of the species were probably contributed directly from the river, surviving, at least for a time, the low-salinity surface waters. Nearest to the river mouths the occurrence in late July of developmental stages of copepods suggests that some others may not have developed in the waters where they were collected. There are several indications of a very low rate of production of zooplankton immediately off the river mouths. Farther off shore, species diversity was greater, in association with "oceanic" water, in which river influence was probably insignificant. Young developing stages of crustaceans as well as an apparently greater biomass point to a higher rate of

production of zooplankton than may occur nearer the fresh waters.

The balance between nutrients, plant and animal plankton is precarious in much of the south Beaufort Sea region, depending upon a varying mixture of river water and offshore influences. The river influence is extensive and variable. It is to be expected that modification beyond the present annual variations in river flow and in the content of river water will change the quantity and composition of the planktonic flora and fauna of a large area of the south Beaufort Sea adjacent to the Mackenzie River delta.

INTRODUCTION

During the last 12 days of July of 1973, the "North Star of Herschel Island" was chartered for an oceanographic cruise in the south Beaufort Sea. Seventeen stations were occupied (Fig. 1) to gather information on water temperature, salinity, dissolved oxygen, nitrate, phosphate, silicate, chlorophylls, particulate and dissolved organic carbon, bacteria and benthic and planktonic plants and animals. The object of the study was to assemble data on the biological oceanography of the Beaufort Sea in the vicinity of the Mackenzie delta, to define selected features of the present marine system. Only by having an understanding of the contemporary, relatively undisturbed state of such an ecosystem can we expect to recognize and appreciate changes in it. Construction taking place in the Mackenzie River may influence conditions downstream from activity sites, to the delta and into the south Beaufort Sea. Knowledge of present conditions in the south Beaufort Sea will permit measurement of changes and estimation of their relevance to the marine ecosystem of the river mouth area. To this end, emphasis in the study was placed on assessment of river contributions to the system and the rather delicate and variable balance which exists between the river influence on the one hand and the offshore marine influence on the other. The sum of these opposing factors seems to determine the pattern of much of the biological structure of the south Beaufort Sea at any one time.

RESUME OF CURRENT STATE OF KNOWLEDGE

The present state of knowledge of the marine ecosystem of the Beaufort Sea is deplorable. This sea, bordering land of both Canada and the United States, has been almost totally ignored by oceanographers until very recently. The first oceanographic paper on the region appears to be Tully's (1952) in which data collected on H.A. Larsen's historic first return journey through the northwest passage on the R.C.M.P. vessel "St. Roche" were given. Tully showed that relatively cold (0.2 - 5.5°C) and brackish (3 - 9‰) water existed between Point Barrow and Liverpool Bay, with warmer and much saltier water (to 31.6‰) to the west, and similarly cold but again more saline (water to 30.7‰) to the east. The relatively brackish water in the middle region was

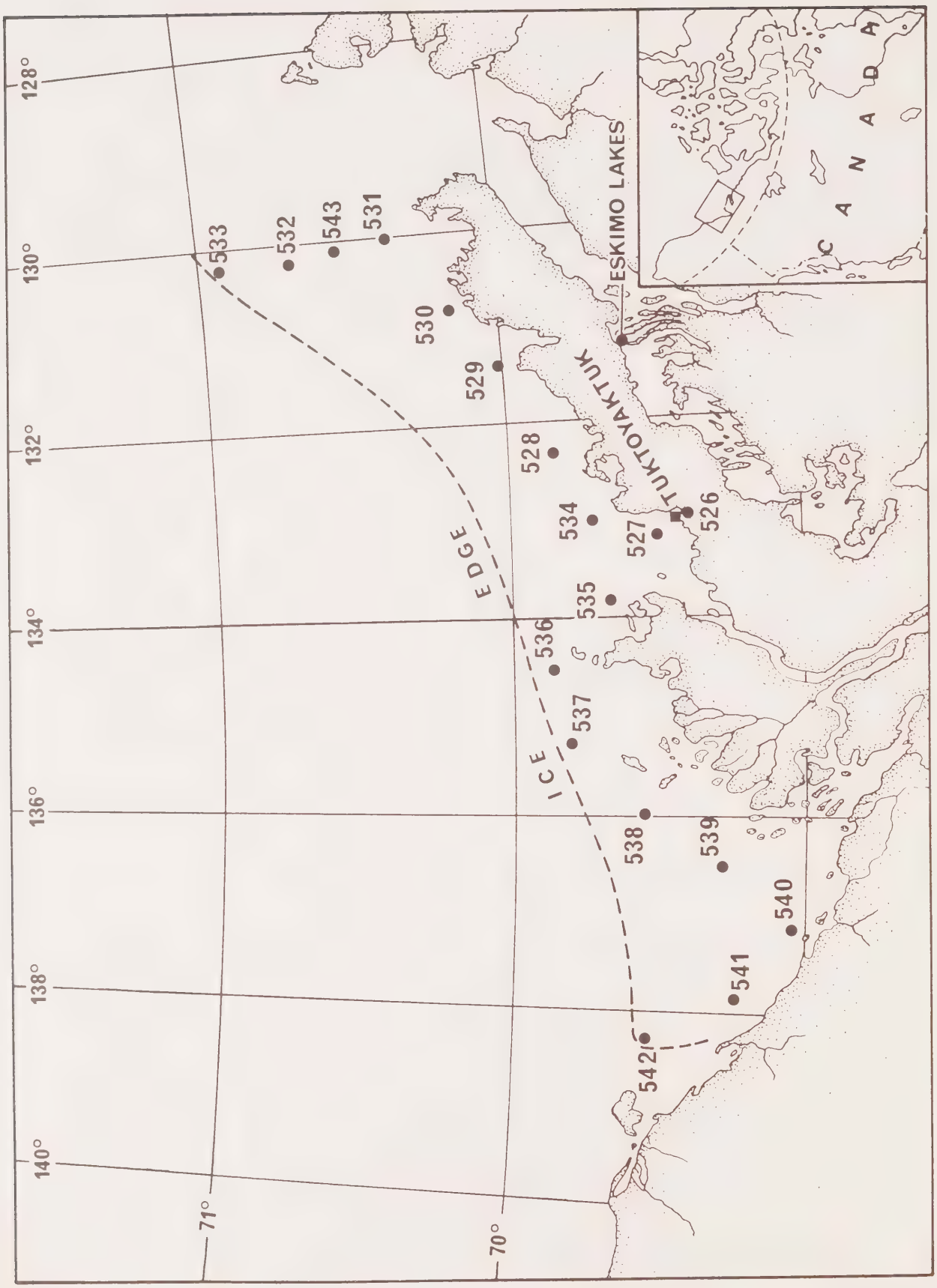

FIGURE 1. STATION LOCATIONS AND THE ICE EDGE, LATE JULY 1973.

of course related to the outflow of the Mackenzie River. Cameron (1953), following two cruises by the Calcolim II in the Beaufort Sea, described the net circulation in the south Beaufort as anticlockwise, with low salinity (Mackenzie) water moving eastward along the coast, and higher salinity water following in behind it from the northwest to mix with river water in Mackenzie Bay. Wind was given as the primary factor in influencing the distribution of low-salinity (Mackenzie) water in the south Beaufort Sea. Coastal salinities relevant to the Mackenzie outflow were offered by Henoch (1969). Detailed inshore oceanography, mainly confined to Tuktoyaktuk harbour, was discussed by Barber (1968). Data from farther off shore, principally on temperature and salinity, were supplied by U.S. Navy Hydrographic Office (1954) and Bailey (1957). Interactions of the Arctic Ocean and the Beaufort and other peripheral seas were discussed by Coachman and Barnes (1961, 1962, 1963).

No published account of nutrients has been found for any part of the south Beaufort Sea, the only truly comparable data so far being from off northern Siberia in the vicinity of the Lena River mouth (Codispoti and Richards 1968). Gudkovich (1955), English (1961) and Kinney, Arhelger and Burrell (1970) have given nutrient data from the Arctic Ocean north of the Beaufort Sea, but their information came from deep, offshore water far distant from the delta coastline. Mackenzie River nutrients are available from Reeder, Hitchon and Levinson (1972).

Published accounts of phytoplankton of the region appear to be limited to Mann's (1925) report on diatoms of the Canadian Arctic Expedition of 1913-18. Later, Bursa (1963) discussed general phytoplankton of north Alaska, to give the only ecological data on these plants in the Beaufort Sea region. Benthic plants (algae) were considered by Collins (1927), and the paucity of data, especially in the western Canadian arctic, was brought out quite recently by Lee (1973). There appear to be no published data on chlorophylls in the south Beaufort Sea. For benthic animals a few scattered collections were made along the north coast of Alaska and closer to the Mackenzie before MacGinitie's (1955) excellent account of the marine invertebrates of north Alaska. The most notable among the earlier works were several papers arising from the Canadian Arctic Expedition, in which a few collections of several taxonomic groups originating in the area of interest here were described. Almost no quantitative data were assembled on the benthic fauna,

however, and no information was available for the assessment of stocks or production rates. Zooplankton was somewhat better covered, and as many as 27 papers pertaining to the Beaufort region were reported by Shih, Figueira and Grainger (1971).

STUDY AREA

The central part of the south Beaufort Sea was chosen as the site for this study, designed to take place off the coast extending from the Mackenzie delta westward to Herschel Island and eastward to Cape Dalhousie. It was intended to have an irregular line of stations along the coast and to run four lines of stations seaward about 70 miles from the near-shore line. At the time of the cruise, however, sea-ice was unfortunately close to the shore (see Fig. 1) and all the outermost stations had to be abandoned as a consequence.

The central part of the south Beaufort Sea is characterized by fairly warm and low-salinity water inshore, and cooler, more saline water offshore. It is in many ways typical of sea areas off large river mouths everywhere, having river-contributed features spreading seaward over deeper oceanic influences below. River flow is dominant in giving the south Beaufort Sea its identity. River flow and varying winds are probably most important in bringing about variations in the distribution of offshore features of the Beaufort Sea in the region of the Mackenzie delta. It is an arctic marine region, and it is ice-bound during much of the year. This means light penetration and vertical mixing are restricted and that production is limited to a fairly brief period of the year. These factors govern to some degree what should be measured in such a system and what the timing of observations should be. The investigations of 1973 were expedient; further work should take better account of the special features of the study area.

METHODS AND SOURCES OF DATA

The samples from sub-surface depths were collected with non-metallic 5-litre Van Dorn bottles or

1.7 litre Niskin bottles. Surface sampling was done with a plastic bucket. Light penetration was measured with a Secchi disc. Dissolved oxygen was analysed in BOD bottles, within a few minutes of collecting, using a YSI model 54 oxygen meter and a magnetic stirrer. Water temperature and salinity were measured *in situ* with a YSI model 33 S-C-T meter. Water samples for chlorophyll were filtered immediately after their collection through HA Millipore filters, which were kept frozen and dark over silica gel until later extraction and analysis of chlorophylls. The remaining water for chemical determinations was kept frozen until later analysis. Phosphate-phosphorus, nitrate-nitrogen, nitrite-nitrogen, silicate-silicon and particulate organic carbon were determined according to the methods of Strickland and Parsons (1968). The slower and far more laborious tasks of plant and animal identifications and counts were conducted (and are still being carried out) under standard laboratory conditions.

RESULTS

1. Physical Oceanography

Data on temperature and salinity are given in Table 1, and the same two features are shown in Fig. 2, and combined in a temperature-salinity diagram in Fig. 3. Highest temperatures and lowest salinities are clearly associated with the Mackenzie River outflow. In Fig. 3, polygon A represents observations made in the upper three metres of nine stations (526, 527 and 534 to 540) closest to river outflow. Polygon C includes observations at the bottom of station 537 and at intermediate depths of stations 541 and 542. Polygon D comprises all observations below 10 metres at stations 532, 533, 541, 542 and 543, the stations farthest from direct river influence. Water from the upper 10 metres at three of the same stations (532, 533 and 543) is shown in polygon E. Polygon B includes all other observations. The pattern is clear and consistent. Warmest and least saline water occurs in the surface layers of the stations closest to the river mouths. The coldest and most saline water is found in sub-surface depths at the stations fartherst from fresh water. Other collection points are intermediate. These observations have particular relevance here because they run parallel to biological features, below.

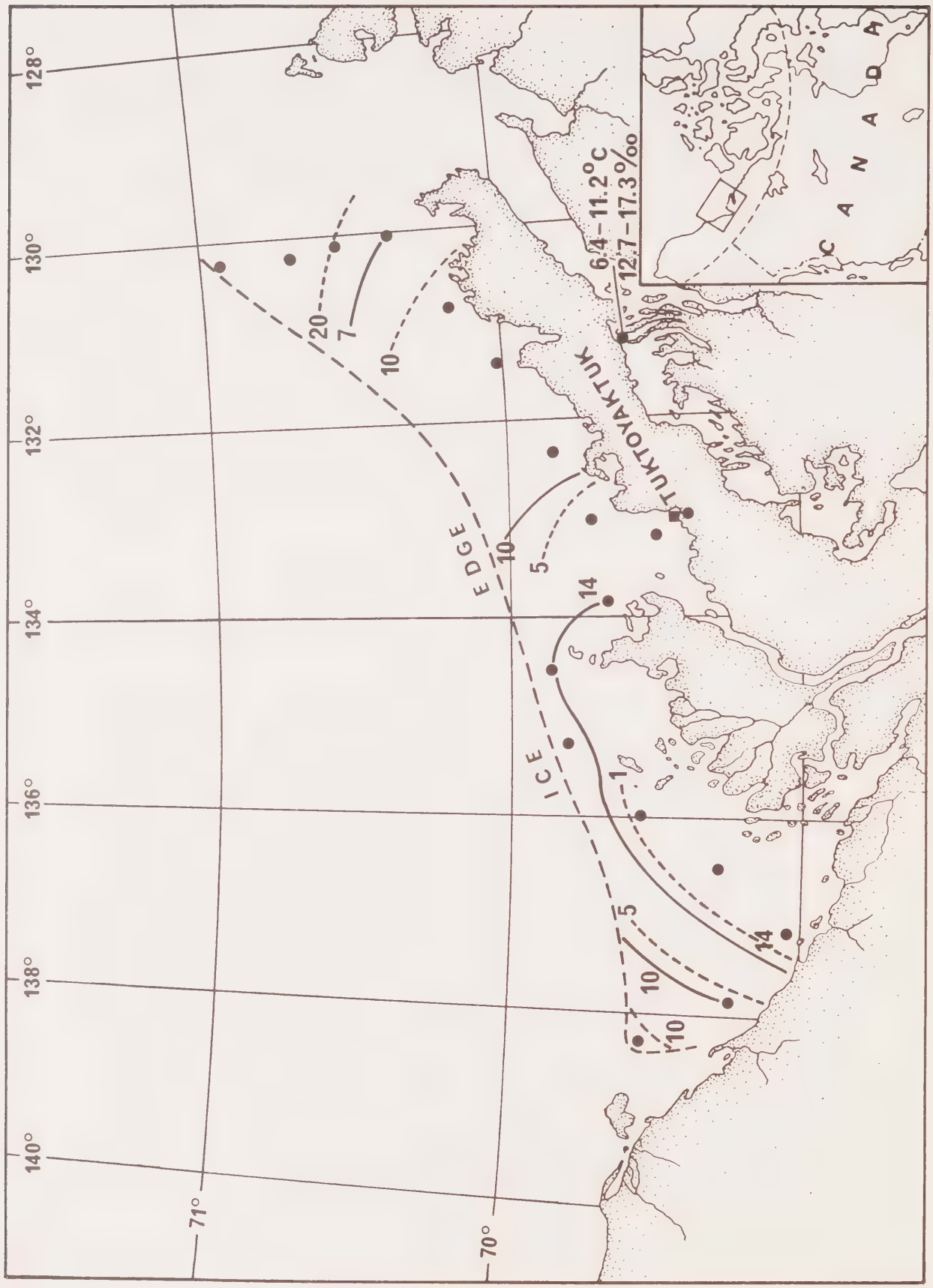

FIGURE 2. TEMPERATURE —— (°C) AND SALINITY --- (°/₀₀) AT THE SURFACE.

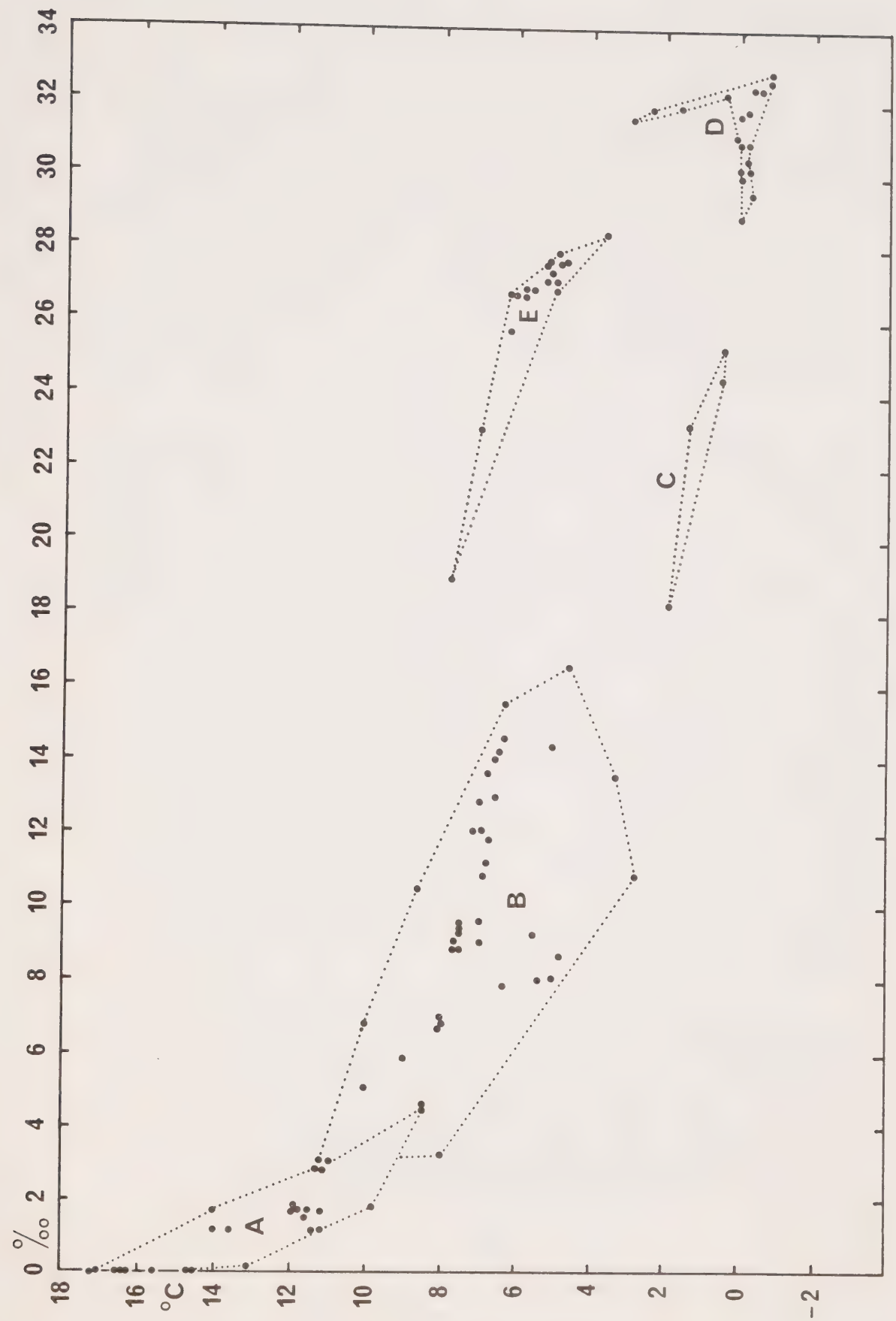

FIGURE 3. TEMPERATURE-SALINITY DIAGRAM. SEE TEXT.

2. Nutrients

Phosphate-phosphorus in the upper five metres (Fig. 4) is distributed in an orderly pattern, with maximal values immediately off the river mouths, and progressively smaller quantities in seaward directions. Profiles (Fig. 5 and 6) show progressively less phosphate in the whole water column as one moves eastward along the Tuktoyaktuk Peninsula. Offshore from Cape Dalhousie, an increase appeared in the deeper water of the two outermost stations, as it did below about 30 metres at the station near Herschel Island. Nearly all phosphate values determined from the south Beaufort Sea were substantially higher than those found at the same depths and during the same period of time in the Eskimo Lakes.

Nitrate-nitrogen in the upper five metres (Fig. 7) also shows an impressively orderly pattern of diminishing values along seaward lines from river mouths. Profiles of nitrate, in the same three sections as were used to show phosphate, illustrate reduction in quantity at all depths in an eastward direction along the coast of Tuktoyaktuk Peninsula (Fig. 8 and 9). Offshore from Cape Dalhousie, all samples from depths less than 20 metres showed no detectable nitrate, while slightly deeper, a high of more than 3 mg-at/mg^3 appeared. Between the Mackenzie River and Herschel Island, high near-river values gave way to smaller quantities near Herschel Island in the upper water layers. In deeper water near Herschel Island, the highest nitrate values of the cruise were found. Nitrates determined at the same time from the Eskimo Lakes were fairly close at all depths to many of the Beaufort stations, with the exception of a few shallow locations close to river mouths where nitrate was considerably more plentiful. This comparison differs interestingly from the one between Beaufort and Eskimo Lakes phosphate, in which all Beaufort Sea phosphate values were higher by a factor of several than those from the Eskimo Lakes.

Nitrite-nitrogen quantities were consistently small, with most samples showing no detectable nitrite, and only two samples with more than 0.1 mg-at/m^3. Positive values were found only at stations 531-533 and at 540-542. Zero readings were made at all depths of all other stations and above 10 metres at station 531, above 10 metres at station 542 and above 20 metres at station 541. Beaufort Sea nitrite values show a range almost identical with that found in the Eskimo Lakes. Alone

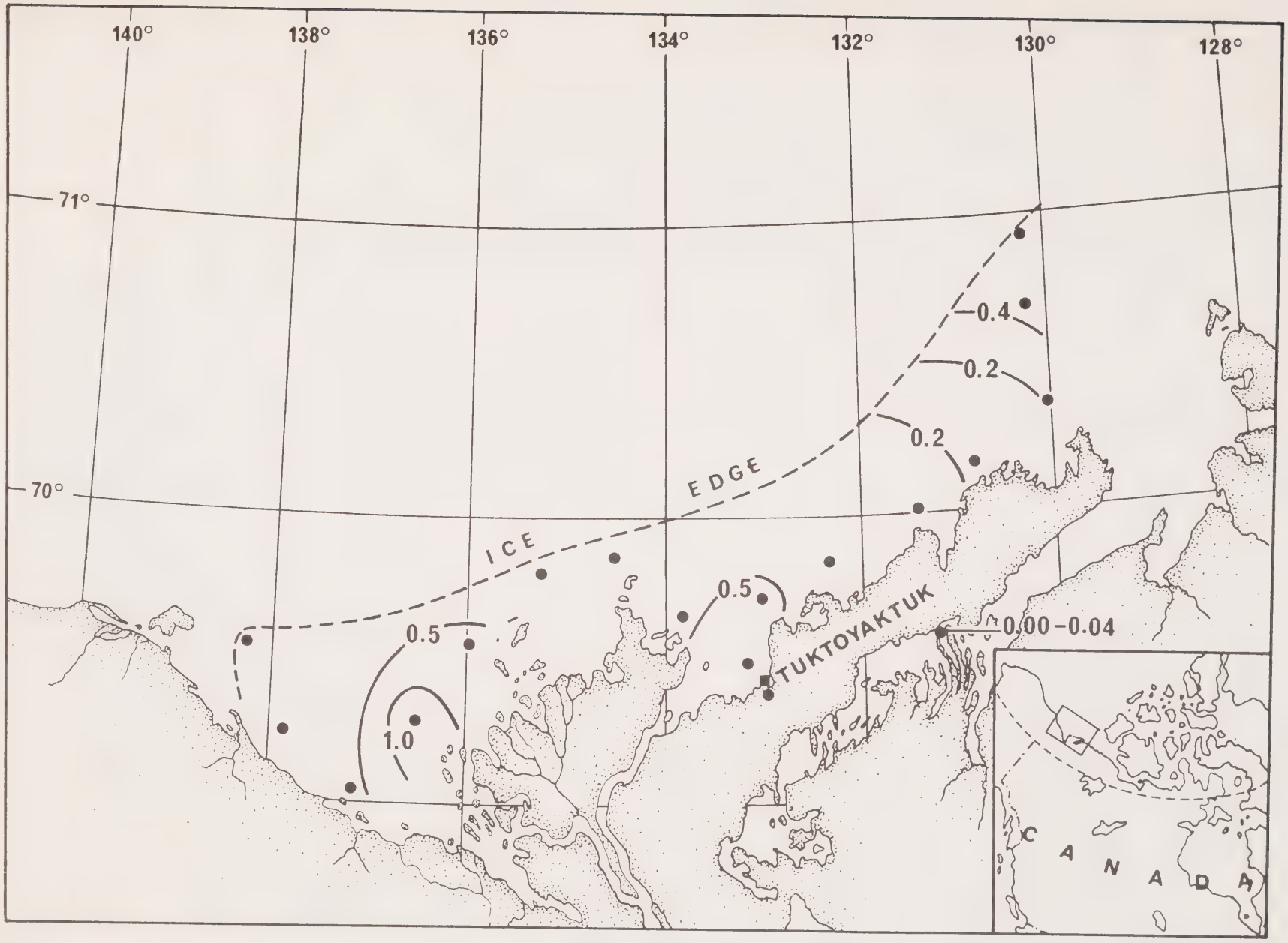

FIGURE 4. PO_4-P (MG-AT/M^3) IN THE UPPER 5 METRES.

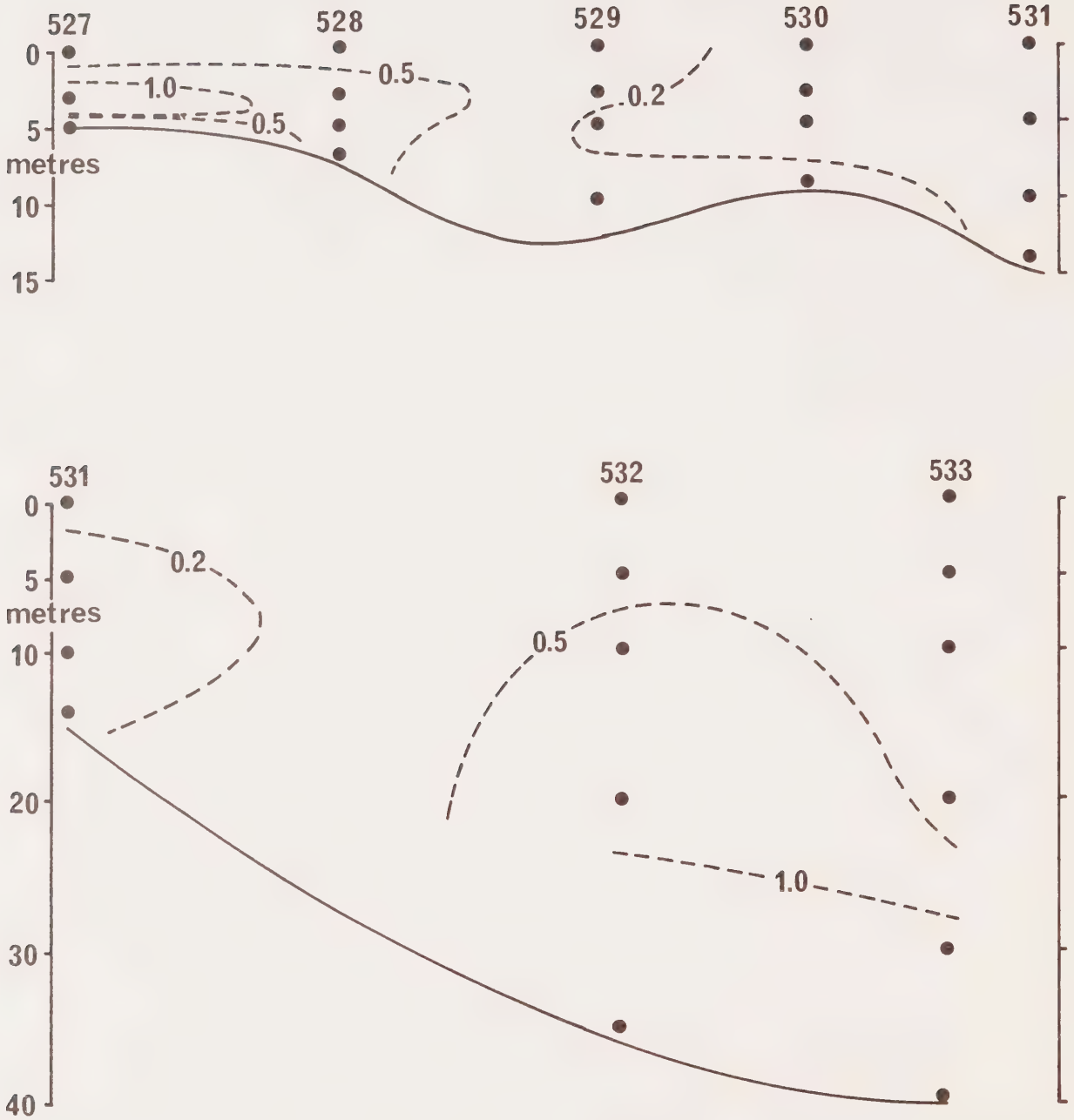

FIGURE 5. PO_4-P (MG-AT/M^3) AT STATIONS 527 TO 533.

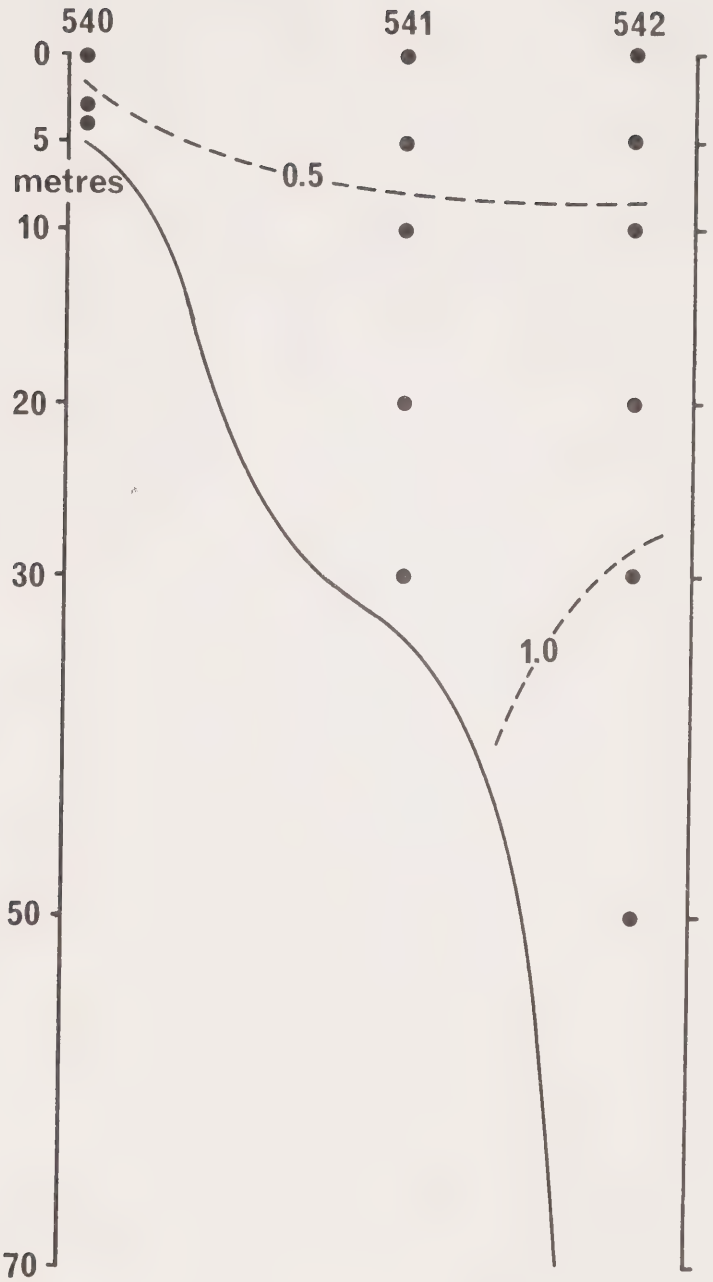

FIGURE 6. PO4-P (MG-AT/M3) AT STATIONS 540 TO 542.

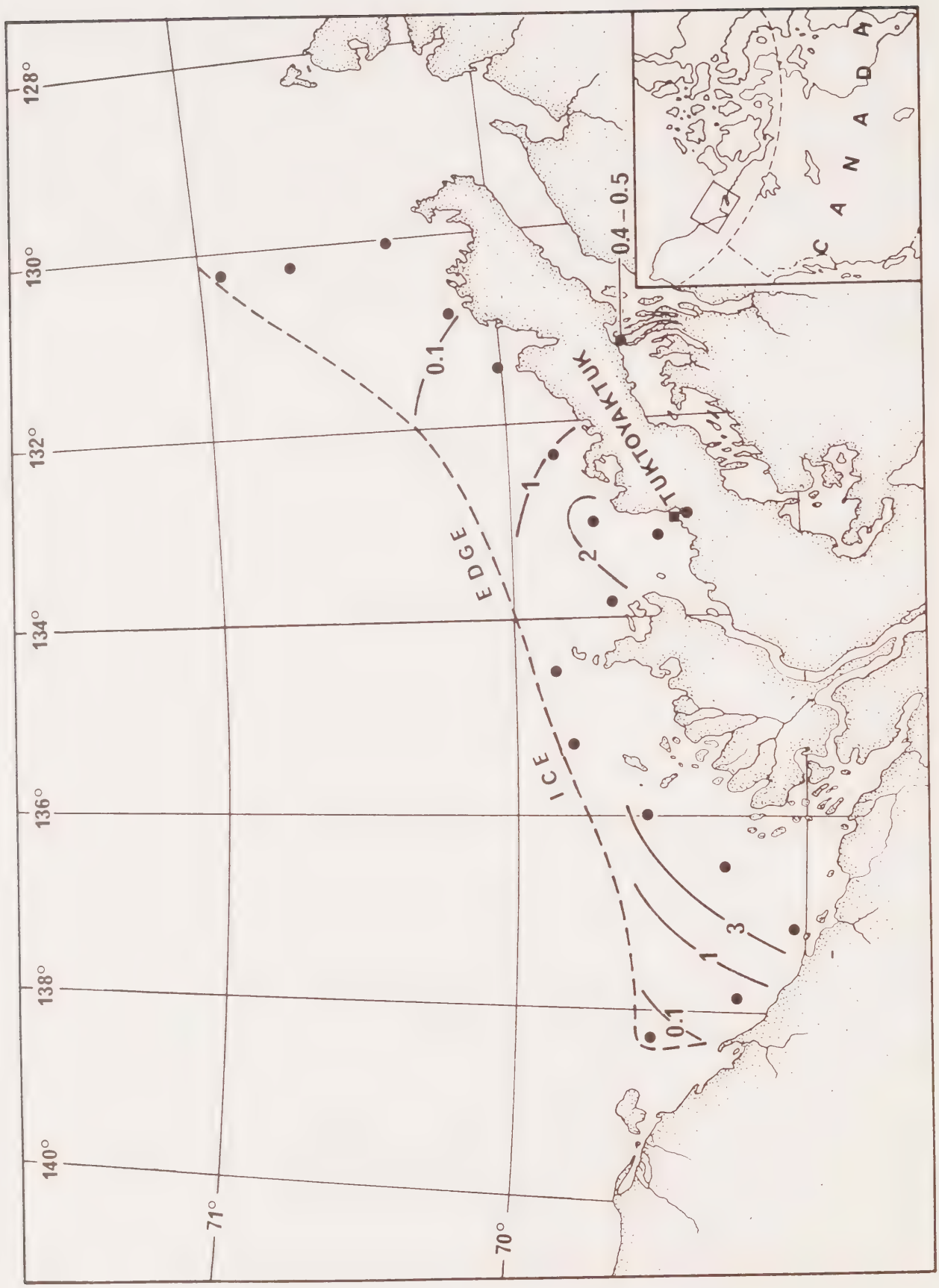

FIGURE 7. NO_3-N (MG-AT/M^3) IN THE UPPER 5 METRES.

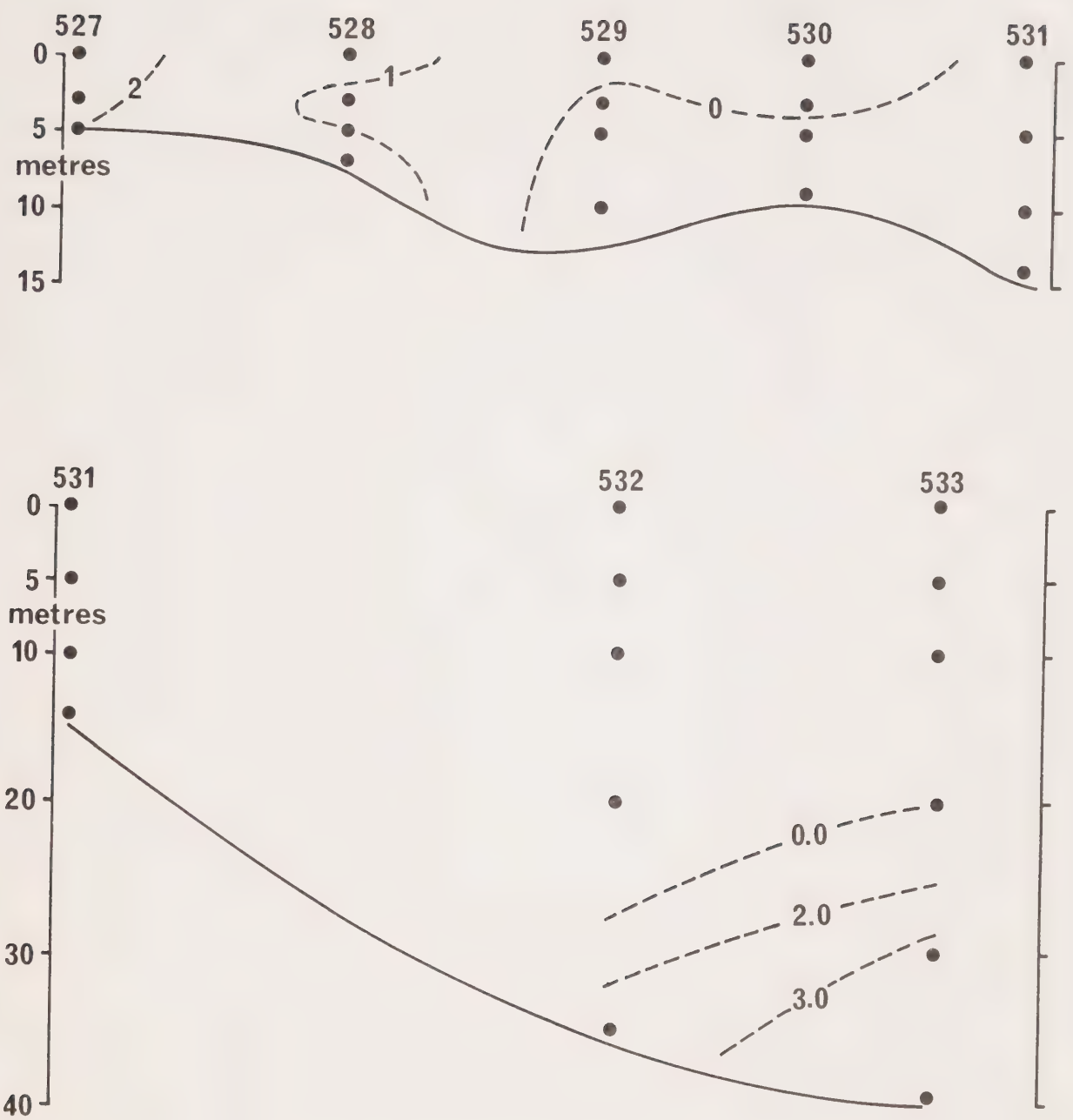

FIGURE 8. NO_3-N (MG-AT/M^3) AT STATIONS 527 TO 533.

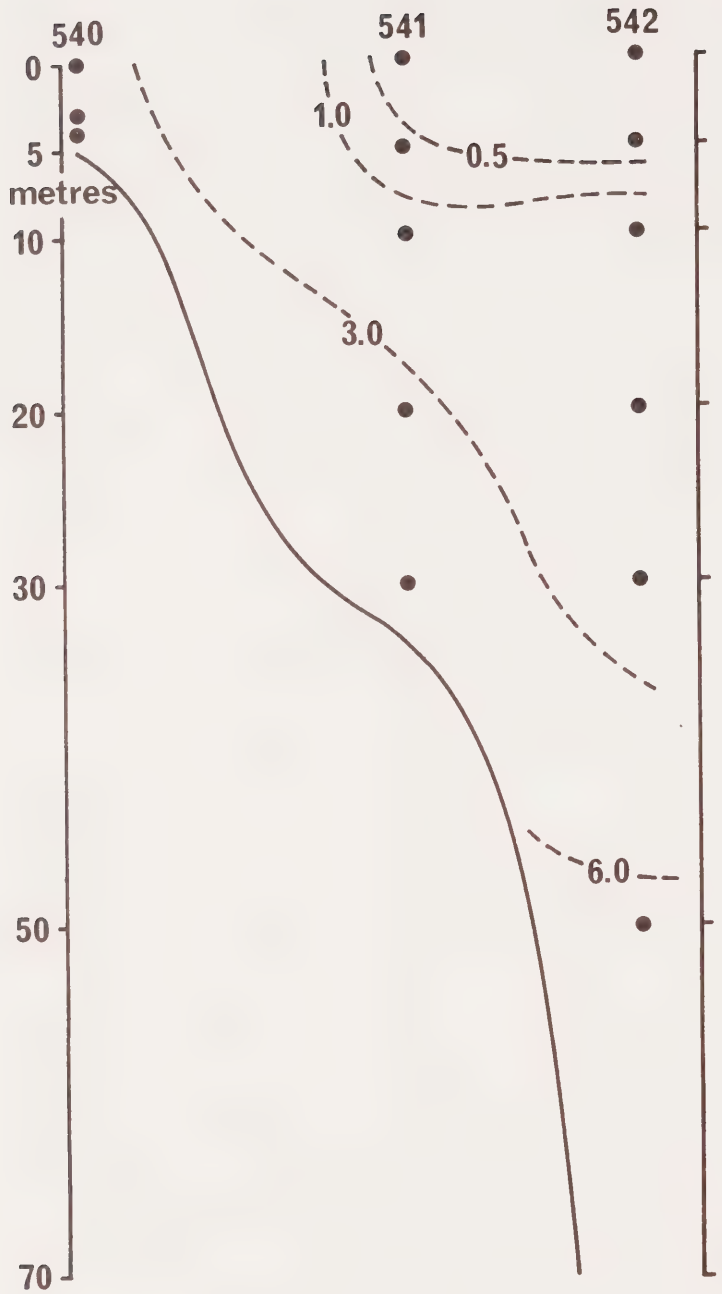

FIGURE 9. NO_3-N (MG-AT/M^3) AT STATIONS 540 TO 542.

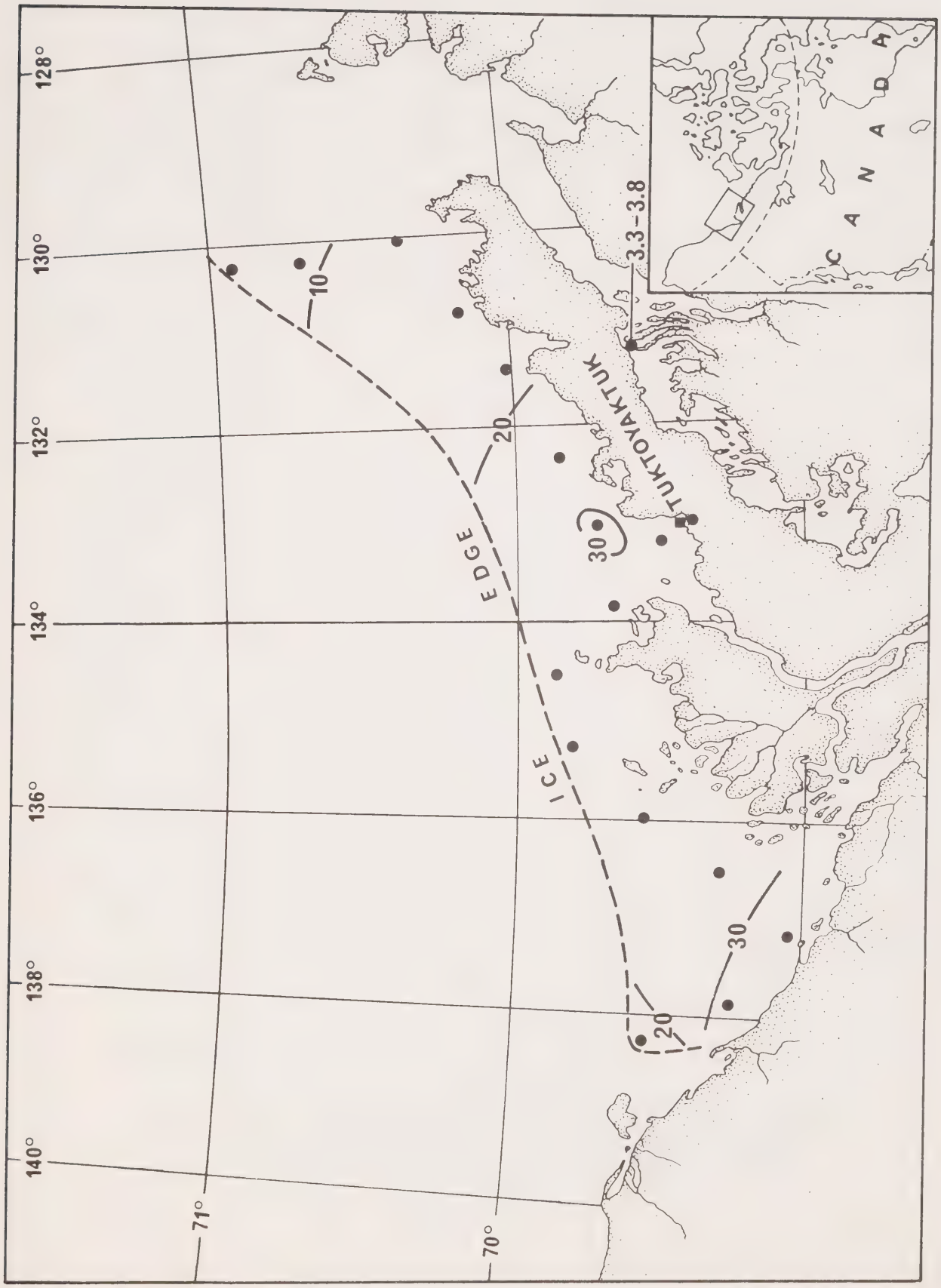

FIGURE 10. Si (MG-AT/M^3) IN THE UPPER 5 METRES.

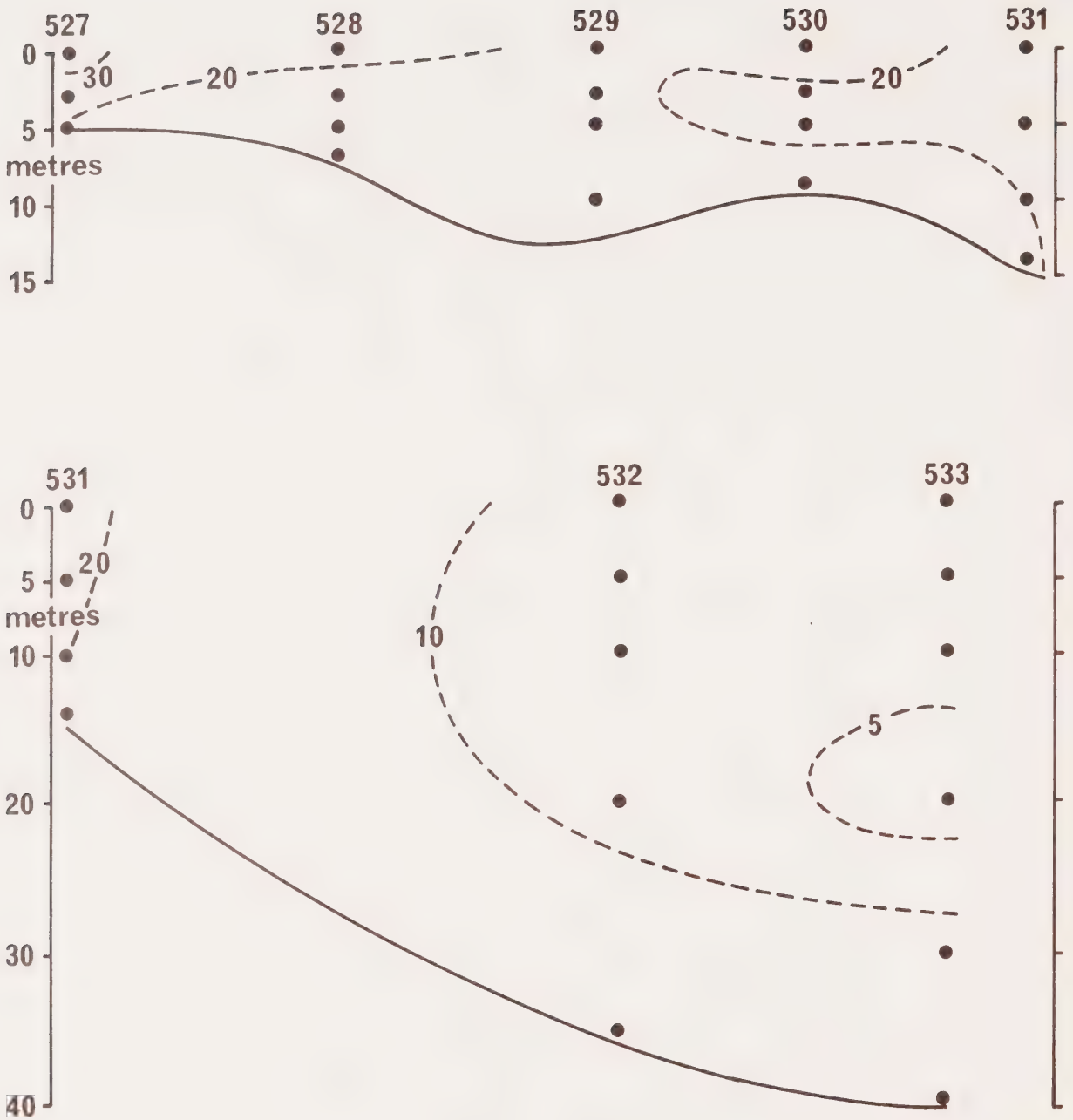

FIGURE 11. SI (MG-AT/M^3) AT STATIONS 527 TO 533.

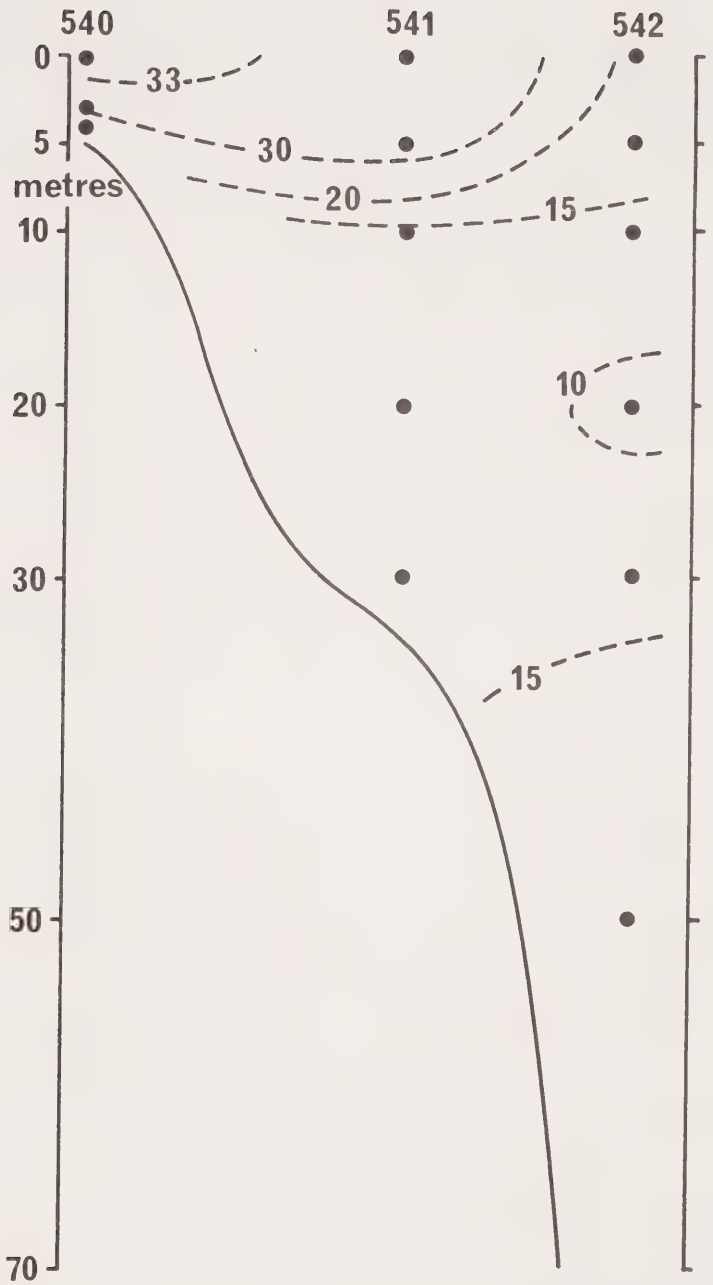

FIGURE 12. SI (MG-AT/M^3) AT STATIONS 540 TO 542.

among the nutrients measured, nitrite showed no concentration at river mouths.

Silicate-silicon (Fig. 10) also shows greatest abundance closest to river mouths. In profile (Fig. 11 and 12), highest inshore and lowest offshore values appear at all depths, and there is not the clear indication evident with phosphate, nitrate and nitrite of relative abundance in deeper, offshore waters. The Eskimo Lakes were low in the upper water layers compared with all the Beaufort Sea stations, but they were comparable at greater depths with values found in the Beaufort Sea.

3. Chlorophyll

The highest values of chlorophyll a (all near the surface) were generally at stations close to discharging river mouths, the lowest at points farthest from the river mouths (Fig. 13). Stations nearest the river had distinctly higher values than were found at the Eskimo Lakes during the same period of time, but those farthest from the rivers had less chlorophyll a.

4. Dissolved Oxygen

Dissolved oxygen was least near the river mouth and most abundant at offshore stations (Fig. 14), and values were close to those at the Eskimo Lakes during the same period of time. In profile (Fig. 15 and 16) oxygen levels are shown to have been highest not only off shore, but well below the surface at the deeper stations. The only subsaturated waters were adjacent to the river mouths, at most depths there. The highest saturation was found at 20 metres at stations 532 and 533, and from 10 to 30 metres at station 542. Saturation values there exceeded 120%.

5. Light Penetration

Secchi disc readings were used to determine the depth of penetration of 1% of the light reaching the surface of the sea. The wide range of values, with a clear pattern of extremely low levels near the shore is shown in Fig. 17.

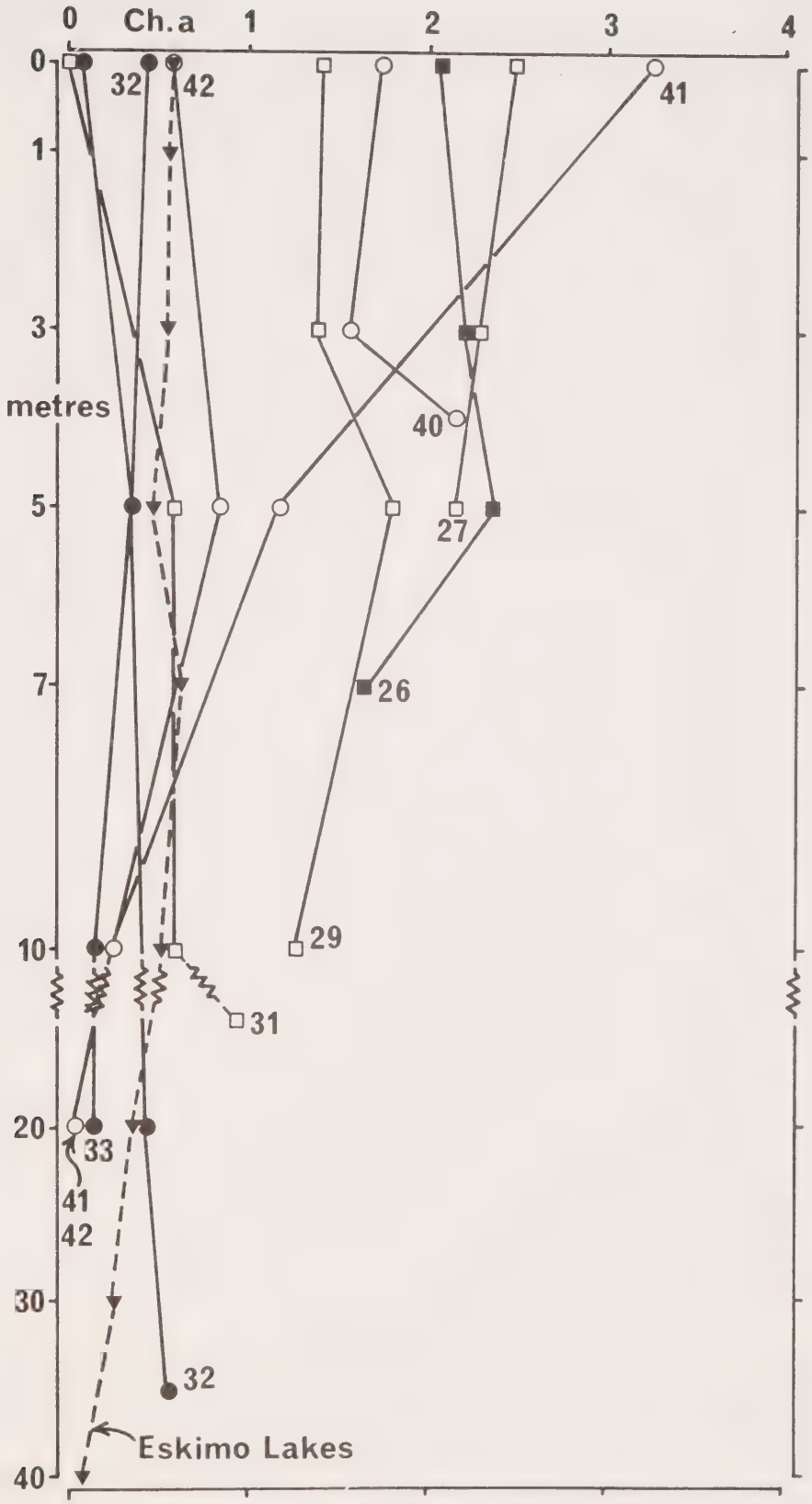

FIGURE 13. CHLOROPHYLL A (MG/M^3) AT SELECTED STATIONS.

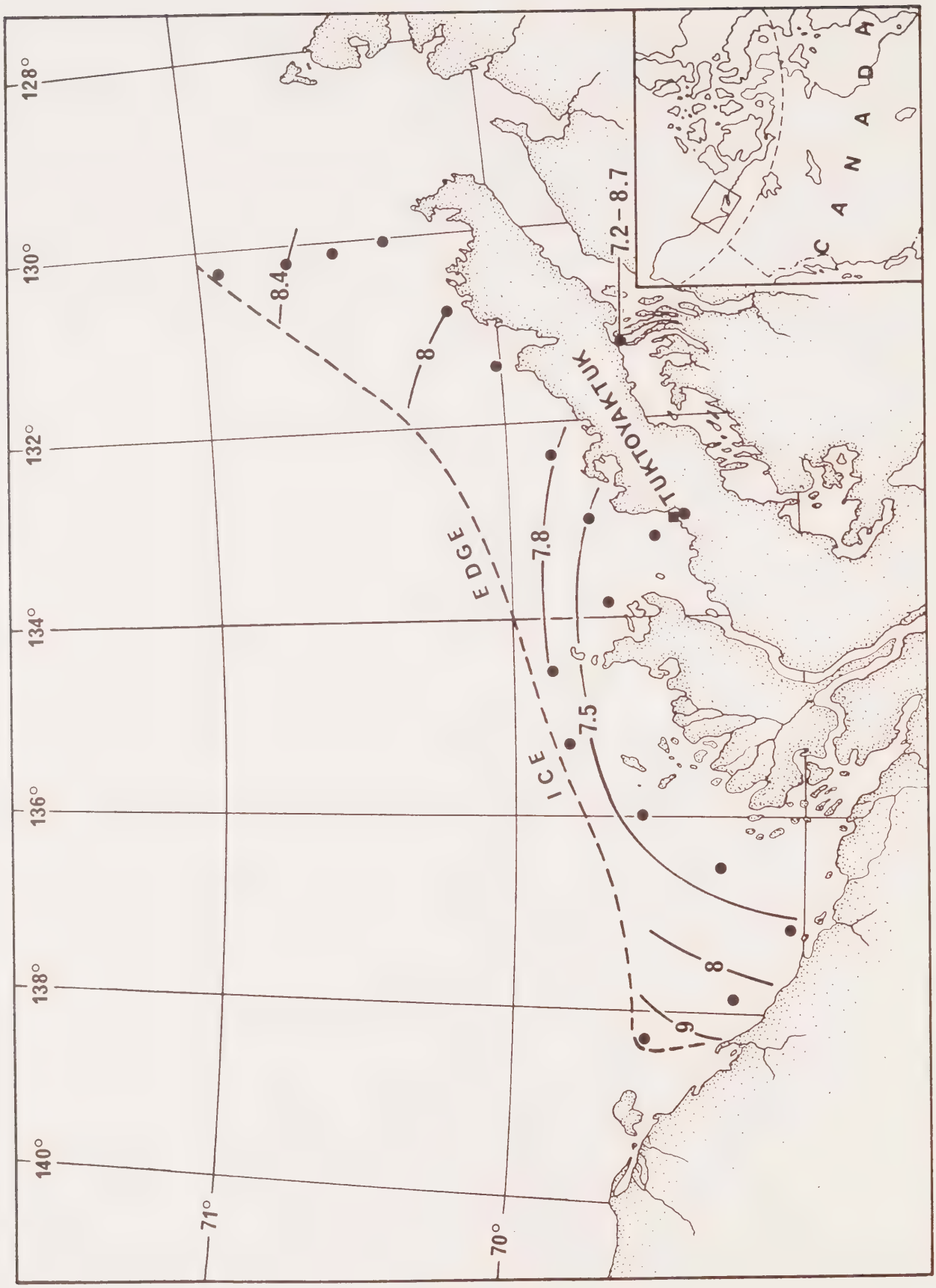

FIGURE 14. DISSOLVED OXYGEN (ML/L) IN THE UPPER 5 METRES.

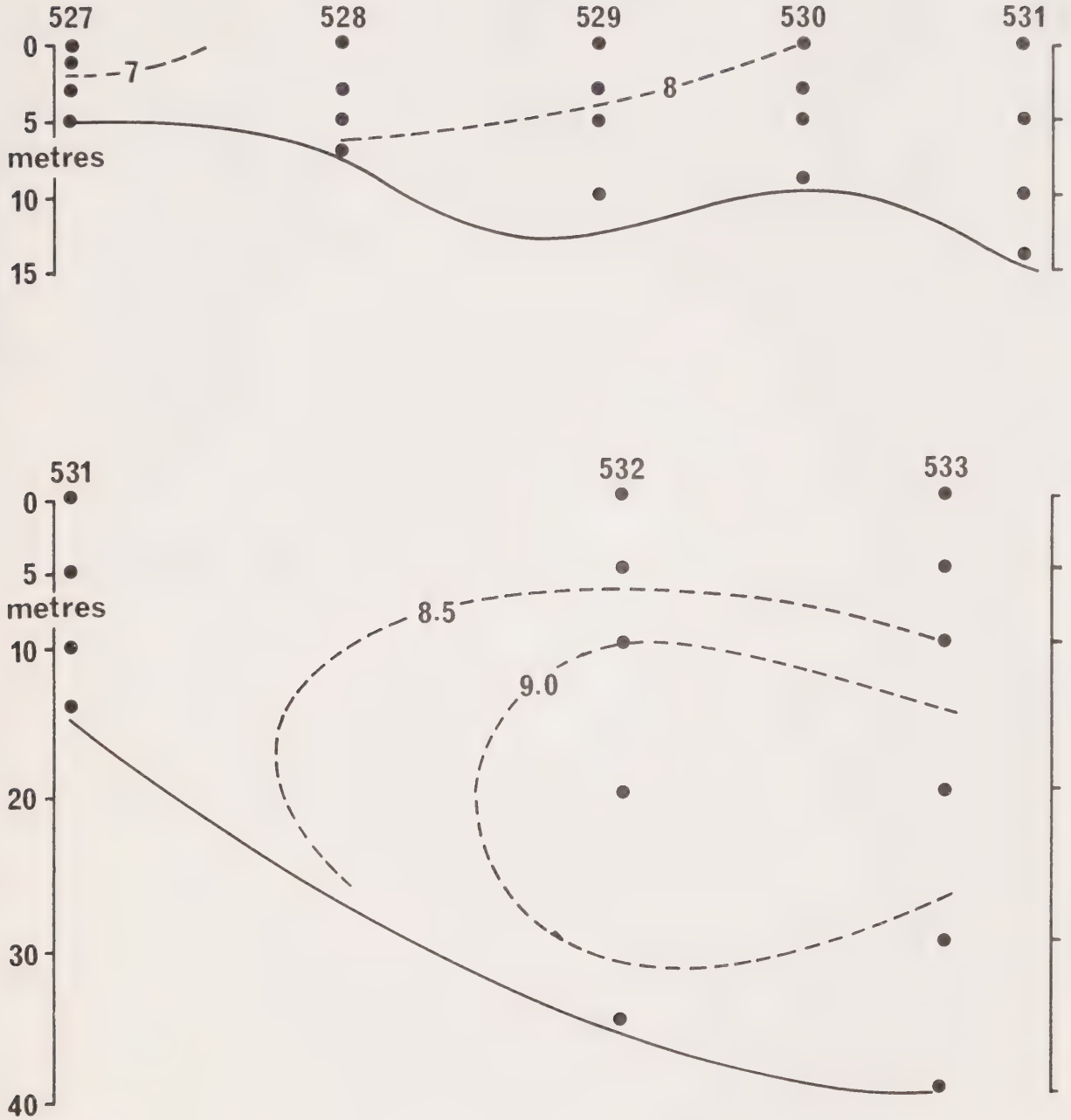

FIGURE 15. DISSOLVED OXYGEN (ML/L) AT STATIONS 527 TO 533.

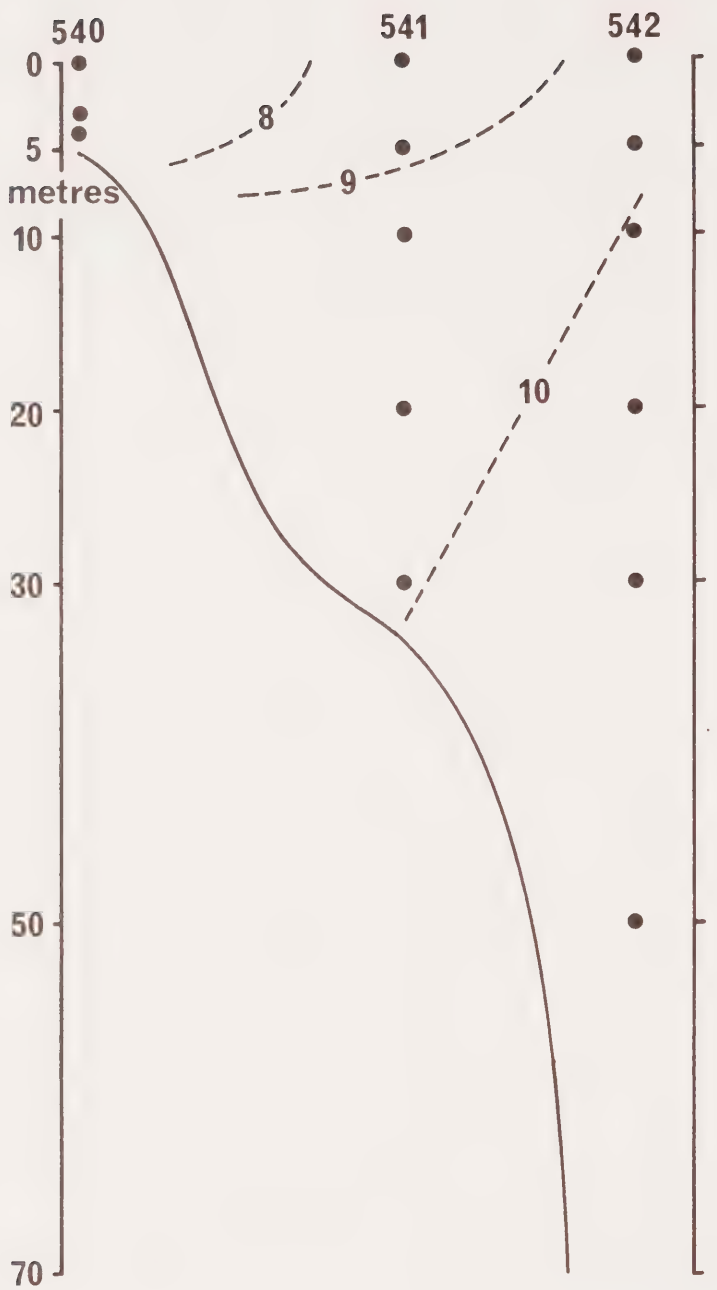

FIGURE 16. DISSOLVED OXYGEN (ML/L) AT STATIONS 540 TO 542.

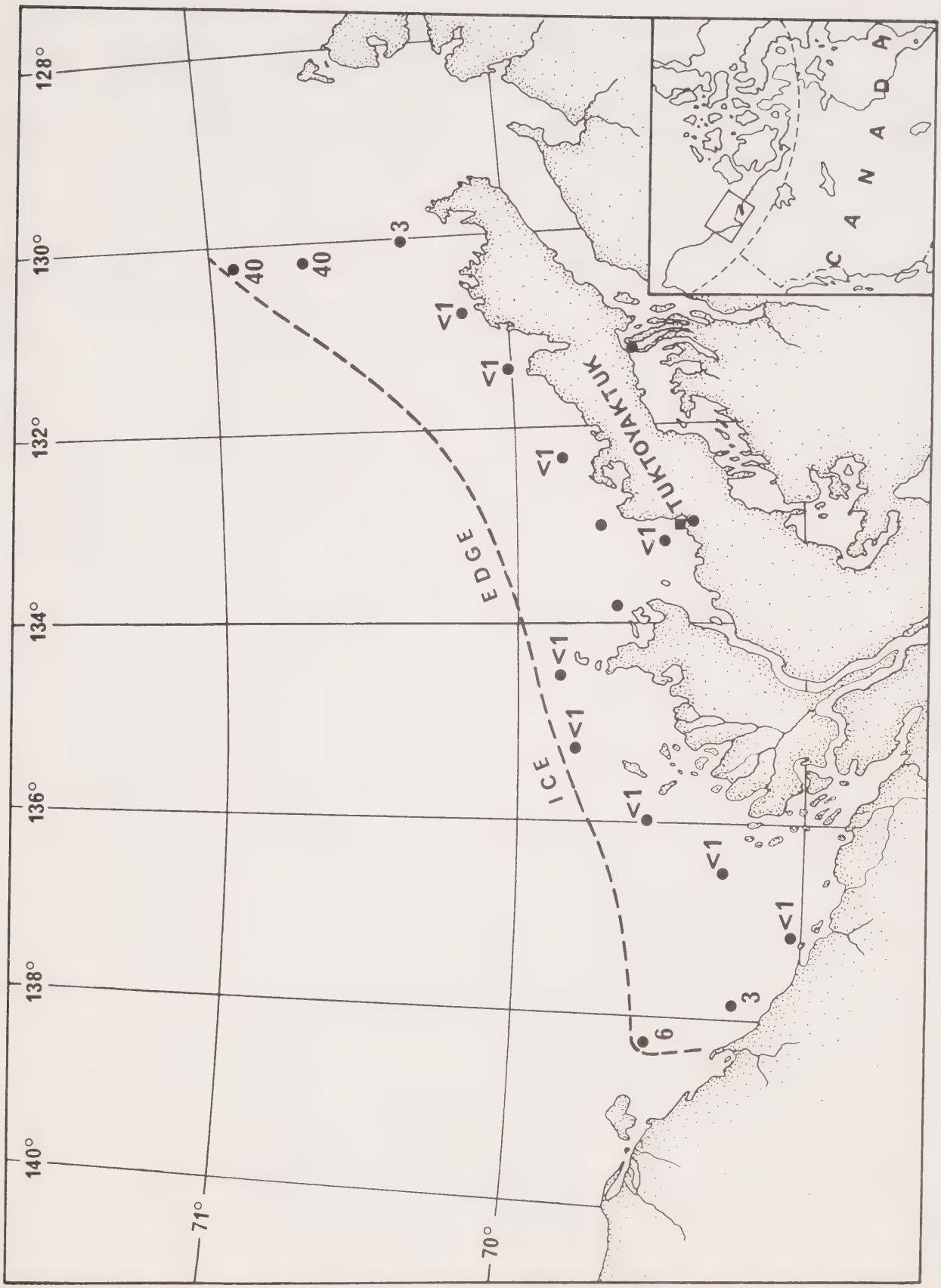

FIGURE 17. DEPTH OF 1% SURFACE LIGHT, IN METRES.

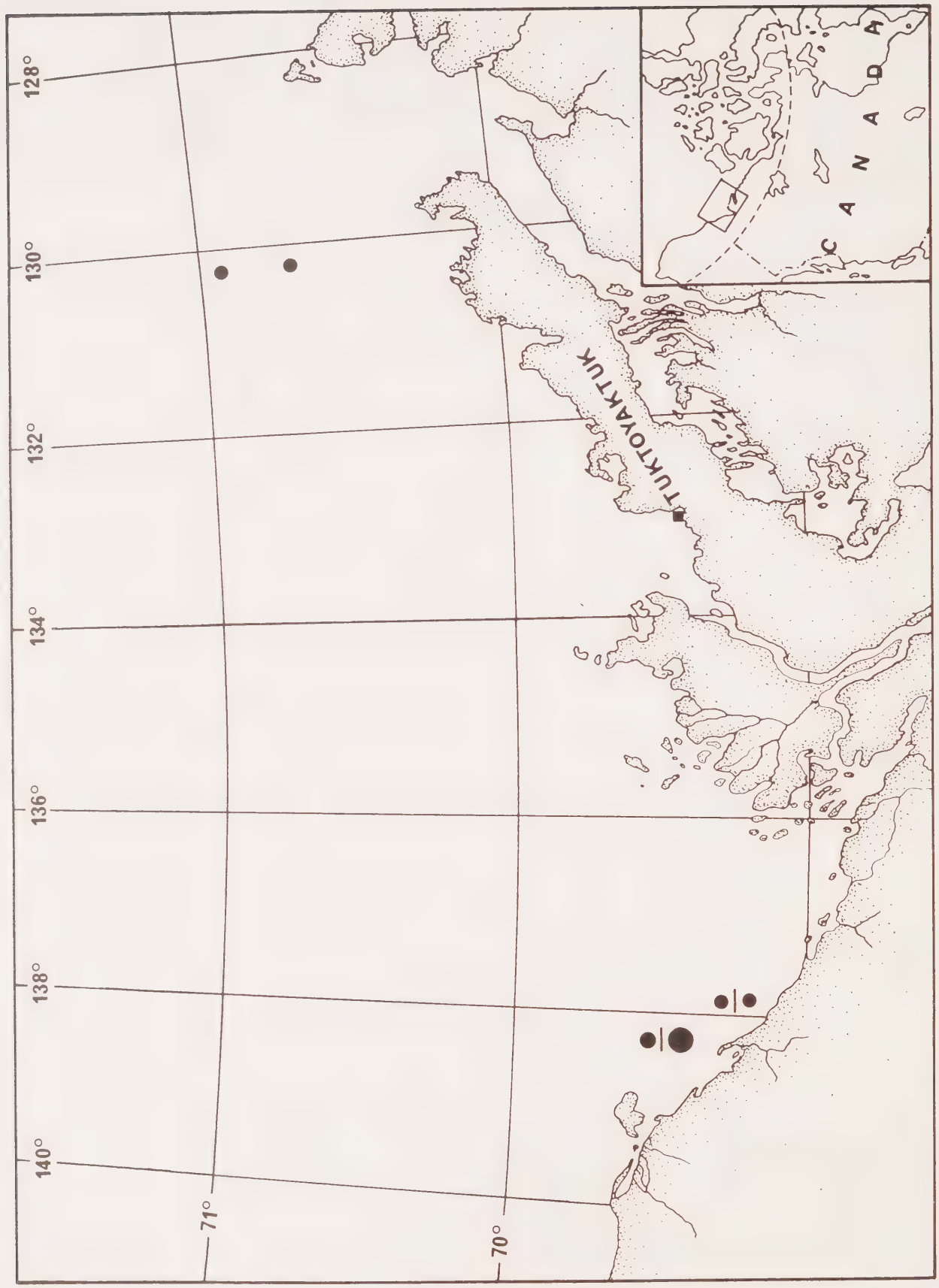

FIGURE 18. OCCURRENCE OF CALANUS SPP. SEE FIG. 19.

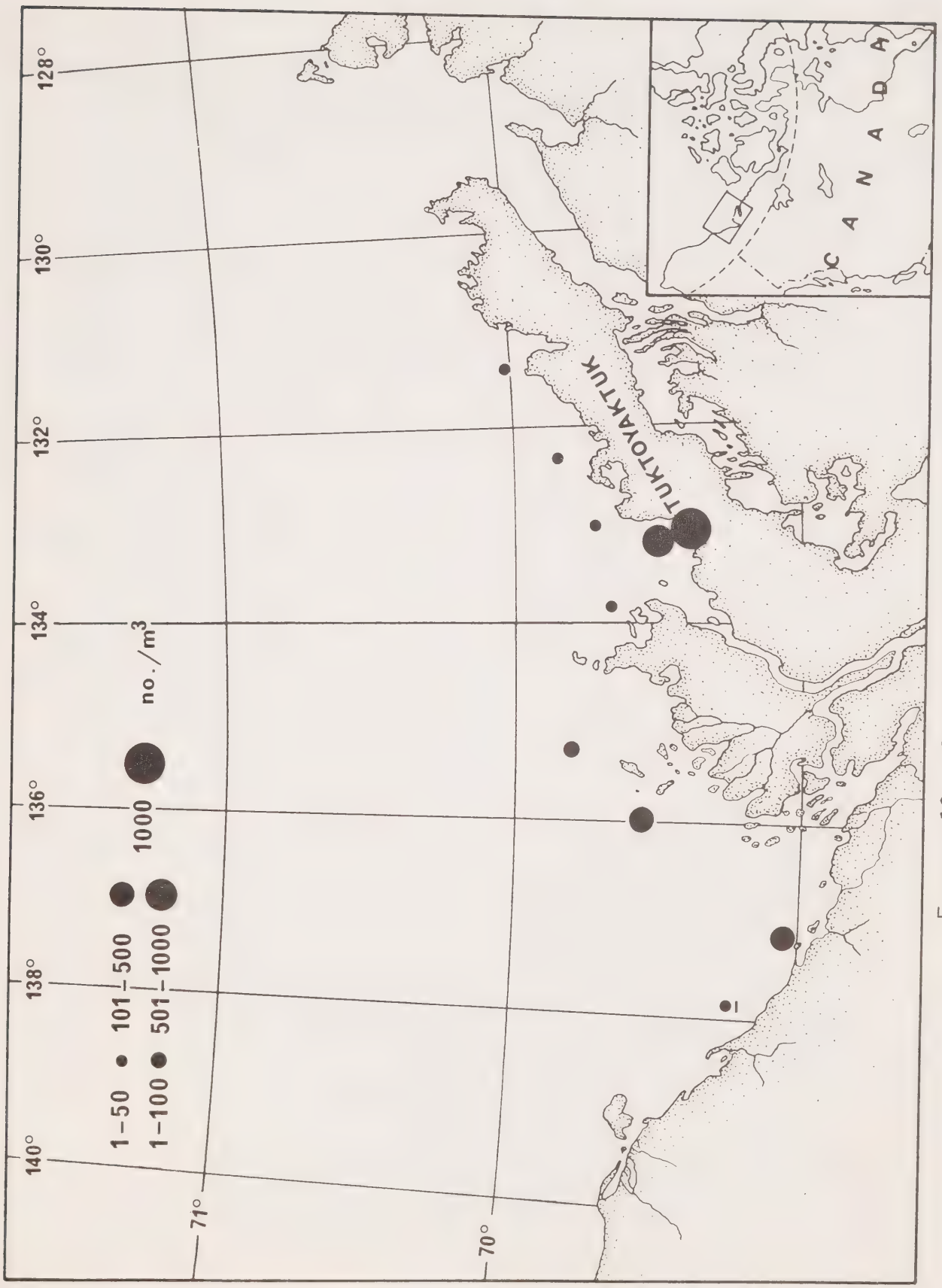

FIGURE 19. OCCURRENCE OF CYCLOPS SP.

6. Zooplankton

Data on zooplankton collections are given in Table 2. At least 45 species were recorded, dominated by 18 copepods. The total number of species may suggest a greater faunal diversity than most of the region in fact supports. At 75% of the stations there were only 13 species found in all. However, at the two stations with the greatest diversity there were 36. The relatively large number of low-diversity stations appear also to have supported a low biomass. They were in the region of maximal Mackenzie River influence during the last week of July 1973, and the few species (crustaceans Cyclops, Limnocalanus, Eurytemora, Mysis and a few others) are characteristic of fresh to moderately brackish water. A few of the species were probably contributed directly from the river, surviving, at least for a time, the low-salinity surface waters. There are several indications of a very low rate of production of zooplankton immediately off the river mouths. Farther off shore, species diversity was greater, in association with "oceanic" water, in which river influence was probably insignificant. There is evidence of a higher rate of production of zooplankton there than may occur nearer the fresh waters.

Most of the species show distribution patterns which indicate dependance mainly upon salinity. Calanus (two species) were all restricted to the stations farthest from the river mouths, those with the highest salinity (Fig. 18). These are characteristically "oceanic" animals, not found in pronounced estuarine conditions. (The sizes of the black symbols in Fig. 18-21 denote numbers of individuals per cubic metre of water samples. See Fig. 19 for explanations. The horizontal bars separating symbols for stations 541 and 542 separate paired symbols for these stations, the upper representing the upper 10 metres, the lower, greater depths at the two stations.) In Fig. 19, the essentially freshwater genus Cyclops is shown, numbers ranging from more than 1,000/m^3 at Tuk to zero at the most remote stations. Eurytemora herdmani and Limnocalanus macrurus are low-salinity species, both, but one rather more than the other, showing preference for river-mouth waters (Fig. 20 and 21).

DISCUSSION

The importance of the Mackenzie River in

determining many of the physical-chemical qualities of the inshore Beaufort Sea is shown. Temperature and salinity distribution indicate relatively warm, low-salinity water flowing outward from river mouths over underlying colder, more saline water. Variations in area and thickness of the overlying river water are not indicated by this single set of observations. It has been suggested from past observations, however, that they have been brought about mainly by changes in wind velocity. Because July of 1973 was a month of fairly quiet weather, without prolonged and constant winds during the weeks immediately preceding the collection of these data, they may be interpreted as indicating a fairly normal condition in the south Beaufort Sea.

Measurements of phosphate-phosphorus are the first reported from these waters. The Russians evidently were the first to give nutrient measurements from the Beaufort-Chukchi area, but from far north of the region of primary interest here. Gudkovich (1955) supplied data on phosphate collected in April between 75 and 80°N. In the upper 50 metres, PO_4-P was found to be consistently close to 1 µg-at/l. Data from Drift Station Alpha (English 1961) from August showed PO_4-P between 0.57 and 1.14 µg-at/l in the upper 50 metres, and from north of the Beaufort Sea in winter (Kinney, Arhelger and Burrell 1970) showed PO_4-P at about 0.8 to 0.9 µg-at/l in the upper 50 metres. In the three reports above, maximum phosphate quantities for the total water columns investigated, ranging from the surface to between 400 and 4,000 metres, varied only from 2.00 to 2.13 µg-at/l and were at either 125 or 150 metres depth.

It is not surprising to find a wider range in the near-surface waters closer to shore, especially in the vicinity of large rivers. Valuable comparative data come from the report by Codispoti and Richards (1968) on nutrients of Siberian coastal waters. There the range of summer values was <0.1 to about 1.5 µg-at/l in the upper two to three metres, and <0.5 to about 3.0 µg-at/l at the bottom. These are higher values than were found in the south Beaufort Sea. In both regions, rivers (the Lena and Mackenzie) were shown to have important effects upon nutrient distribution, highest values occurring in proximity to the river mouths.

There is some information available on phosphate present in fresh waters in the Mackenzie delta region. Thirty-six samples from various rivers, streams and tundra

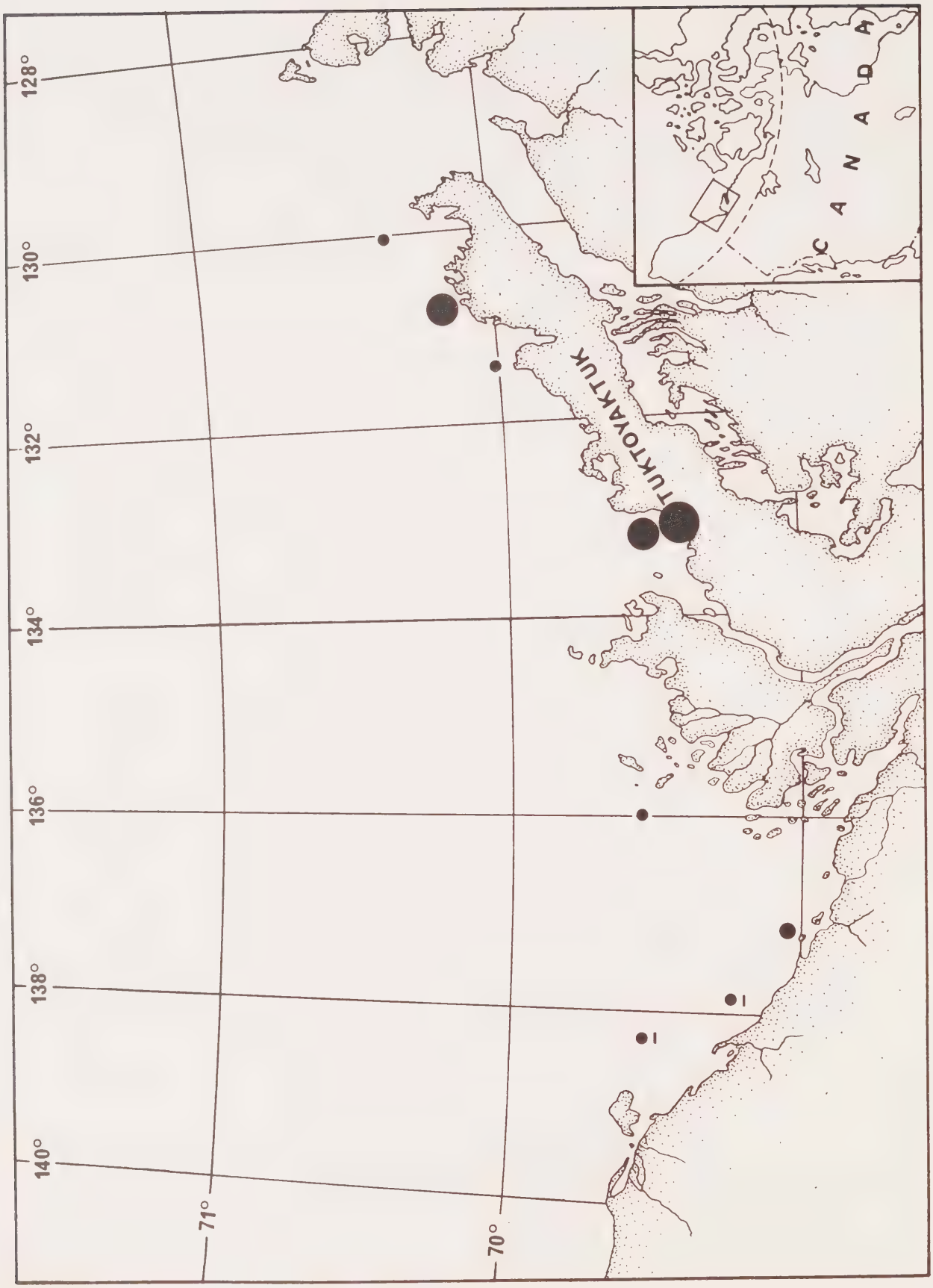

FIGURE 20. OCCURRENCE OF EURYTEMORA HERDMANI. SEE FIG. 19.

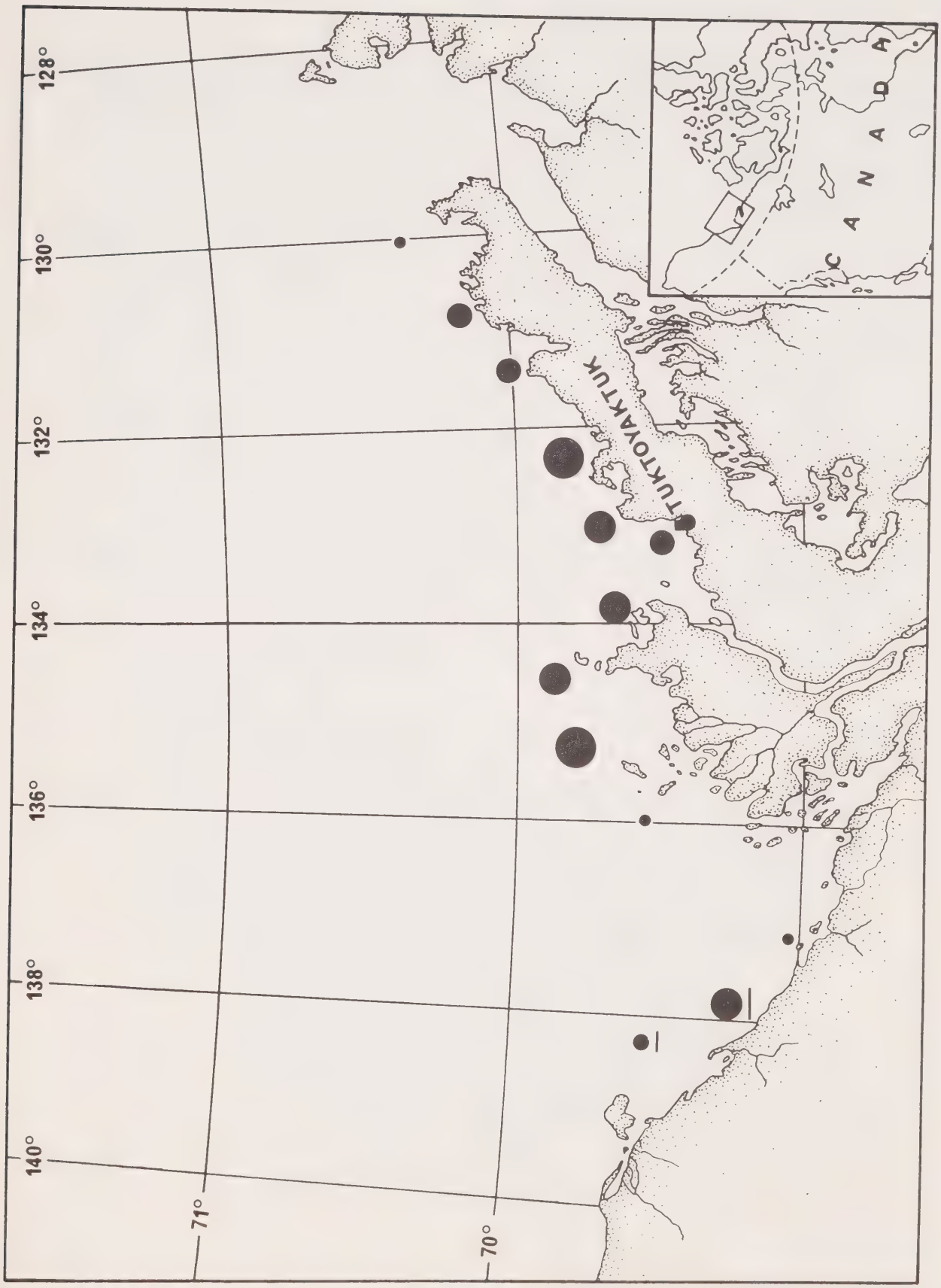

FIGURE 21. OCCURRENCE OF LIMNOCALANUS MACRURUS. SEE FIG. 19.

ponds mainly south and east of the Eskimo Lakes and Liverpool Bay were analysed for various components including PO_4-P in late July of 1973. The range was 0 to 0.39 µg-at/l. Reeder, Hitchon and Levinson (1972) found, from 101 stations in the Mackenzie drainage area, a PO_4 range of <0.01 - 0.09 mg/l (= PO_4-P of <0.10 - 0.95 µg-at/l), and from a single station in the Mackenzie River where it enters the delta, and from where the river's contribution to the sea may best be assessed, <0.01 mg/l PO_4 (= <0.10 µg-at/l PO_4-P). All these are significantly lower than the highs of nearly 1.5 µg-at/l PO_4-P found near the surface in Mackenzie Bay and off the east branch of the Mackenzie River, and in sub-surface water at the stations farthest from shore.

Nitrate-nitrogen values were also formerly unknown from inshore Beaufort waters. Gudkovich (1955) found April quantities between 0.7 and 1.4 µg-at/l in the upper 50 metres north of the Beaufort-Chukchi region, English (1961) a range of 0.4 to 1.7 µg-at/l in the upper 50 metres at station Alpha in August, and Kinney, Arhelger and Burrell (1970) about 1.0 µg-at/l in the upper 50 metres in winter in the north Beaufort Sea. Highest nitrate quantities found in the total water columns sampled by the authors above were between about 11 and 17 µg-at/l, between 150 and 200 metres down in columns ranging in depth from 400 to 4000 metres.

As with phosphate, a wider range of values may occur nearer shore, in proximity to river mouths, as shown by Codispoti and Richards (1968) on the Siberian coast. There, in summer, they reported a range of <0.5 to >2 µg-at/l NO_3-N in the upper five metres and bottom values of <1 to >15 µg-at/l. Both surface and bottom amounts were greatest at stations closest to the Lena delta. Values were higher than those found off the Mackenzie delta (range of the latter, 0 - 9.5 µg-at/l), but showed the same kind of distribution pattern with respect to the river mouth.

Nitrate was found in rivers, streams and tundra ponds south and east of the Eskimo Lakes in late July to be from 0 to 0.7 µg-at/l. Reeder, Hitchon and Leninson (1972) reported a range of values in the Mackenzie drainage area of <0.01 - 0.77 mg/l NO_3 (= <0.2 - 12.4 µg-at/l NO_3-N), and in the Mackenzie where it enters the delta 0.28 mg/l NO_3 (= 4.5 µg-at/l NO_3-N). These are

all lower quantities than the maximum found in the south Beaufort Sea, where the highest levels were found near the bottom at the stations farthest off shore.

Nitrite from the south Beaufort Sea varied in quantity between 0 and 0.14 µg-at/l NO_2-N. Gudkovich (1955) found 0.04 - 0.21 µg-at/l, and English (1961) 0.01 - 0.08 µg-at/l in the upper 50 metres north of the Beaufort Sea. Rivers, streams and tundra ponds south and east of the Eskimo Lakes showed a range of 0 - 0.41 µg-at/l in late July, and these appear to be the only data available from nearby terrestrial sources.

Silicate-silicon quantities in April reported by Gudkovich (1955) were 11 to 18 µg-at/l in the upper 50 metres. The data from English (1961) gave a range of 3 to 9, and from Kinney, Arhelger and Burrell (1970) 5 to 10 µg-at/l in the upper 50 metres. Highest values of all depths at these locations were found between 27 and 57 µg-at/l, at 150 and 180 metres. Nearer shore, off the Siberian coast in summer, Codispoti and Richards (1968) found a range of <10 to >30 µg-at/l in the upper 5 metres, and of <10 to >50 at the bottom, with highest quantities at all depths being found closest to the Lena River delta. These levels were very close to what was found in the south Beaufort Sea, 2.8 to 38.8 µg-at/l at all depths.

Silicate found in the rivers, streams and tundra ponds south and east of the Eskimo Lakes ranged from about 7 to 62 µg-at/l. Reeder, Hitchon and Levinson (1972) found a range of 0.3 - 7.6 mg/l SiO_2 (= 5 - 127 µg-at/l Si) in the Mackenzie River drainage area, and a value of 3.5 mg/l SiO_2 (= 58 µg-at/l Si) in the Mackenzie River where it joins the delta. It is perhaps important that the quantities given here from terrestrial sources are considerably higher than those found in the adjacent sea, which is quite different from what was found with other nutrients, which were more plentiful in the sea than in contributing rivers.

In late July the quantity of chlorophyll a in the Eskimo Lakes was about 50% of maximum values which were found at the end of June. Because ice break-up off the mouths of the Mackenzie took place at about the same time as it did in the Eskimo Lakes, it is to be expected that the period of highest quantity of chlorophyll a reflecting in situ photosynthetic activity occurred at close to the same time in both locations in 1973. In

late July, however, chlorophyll levels at the inshore stations were too high to have declined from a reasonable maximum a few weeks earlier. At the same time, levels at stations farther off shore were low, similar to contemporary conditions in the Eskimo Lakes, and reasonable residues of a phytoplankton bloom a few weeks earlier. The most plausible conclusions which can be drawn from conditions at the inshore stations are either that there is sustained production just off the river mouths, or that there is a constant replenishment of chlorophyll from the outflowing river.

The low level of inshore oxygen suggests an extremely low rate of plant production, on the basis of comparison with conditions in the Eskimo Lakes. This is in contrast with the higher offshore quantities, which are closer to the level expected in waters supporting even a fairly low rate of primary production. Oxygen levels at the river mouths at the sampling time therefore do not support the suggestion of sustained plant production of any consequence immediately off the river, leaving river contribution at the best explanation for the moderately high chlorophyll a readings there.

Nearly all the inorganic nutrients measured, phosphate, nitrate, silicate, showed highest surface values at river mouths, lowest at stations farthest off shore. These are characteristic features for phosphate, nitrate and silicate of estuaries and off large rivers, and according to Emery and Stevenson (1957, p. 693): "The relatively high concentration of nutrients in estuaries and lagoons is evidently the result of nearness to the supply provided by the land runoff and the continuous replacement of nutrient-rich water to growing plants." Phosphate and nitrate show another set of high values, in the deep water of stations 532, 533 and 542. There is less evidence of this in the distribution of silicate.

Chlorophyll a is most abundant at the surface near the river mouths, least plentiful off shore at the surface and in deeper waters. This condition (at the surface) was found by Riley (1937) off the Mississippi River mouth and by Ketchum (1967) in New York Bight, off the mouths of the Hudson and Raritan Rivers.

According to Ketchum (1967), estuaries may be fertilized by (1) river waters leaching plant nutrients from soil and carrying them to the estuary, (2) pollution, contributing extra nutrients, and (3) subsurface

counter currents delivering deep nutrients from below the euphotic zone to the surface. The first of these appears to be clearly applicable to the south Beaufort Sea, and regarding nutrients there, there seem to be at least two significant questions. Do high nutrient levels at river mouths reflect high, continuing supply from rivers, low utilization of the supply in coastal waters, or both? And, what factor or factors function to limit coastal production if nutrients are in strong and continuing supply?

Oxygen and chlorophyll data led to the conclusion (above) that the primary production rate, at least during the early part of summer, was low in the waters immediately off the river mouths in the south Beaufort Sea. A low primary production rate of course will not put a strong demand upon available nutrients. It is suggested that high nutrient levels at the river mouths reflect a continuing supply from the rivers, a supply which is not used in any large measure by plant plankton in these waters.

Nutrients are evidently not limiting at most of the south Beaufort stations, but light probably is. Fig. 17 shows the depth of penetration of 1% of surface light. The intense turbidity is clearly shown by the values. At all stations, except those farthest from the river mouths, light penetration was almost totally inhibited by the suspended matter in the water. Primary production in such waters is restricted to the upper few centimetres, and can be only very small under these circumstances. Only at stations 532 and 533 can production be expected to be reasonably high on the basis of available light for photosynthesis. In fact it was not so at the time these collections were made, the phytoplankton bloom probably having occurred earlier in the summer. Nitrate was exhausted in the upper waters, and chlorophyll *a* was very low.

CONCLUSIONS

1. The Beaufort Sea adjacent to the Mackenzie delta shows estuarine properties, with outflowing river water moving seaward from the various river mouths over an inward extension of marine water below.

2. Nutrients are contributed to the surface waters of the sea from the outflowing Mackenzie River, and standing values of PO_4-P, NO_3-N and Si near the delta, at least in late July, are high.

3. Phytoplankton appears to be more abundant just off the delta than farther off shore. Values immediately off the river mouths probably reflect mainly river transported plant material.

4. Primary production appears to be low adjacent to the delta, in spite of plentiful nutrients, probably largely because of the high turbidity of the water which reduces light available for photosynthesis.

5. Primary production farther off shore is probably generally higher than nearer the shore, during ice-free periods, because of relatively uninhibited light penetration and availability of nutrients from sub-surface depths.

6. Secondary production appears to be fairly low in the delta region, and to be greater in the clearer waters farther from shore.

RECOMMENDATIONS

The Beaufort Sea is a sump to the Mackenzie River. Certain events occurring in the river inevitably will be reflected subsequently in the Beaufort Sea. The most important forms of impact on the river which would influence Beaufort Sea conditions would be changes in river flow and in the composition of the river water. The second of these, changes in water composition, could follow upon additions of particulate matter or dissolved substances to the river. The extent of influence in the Beaufort Sea will depend upon the quantities of additives and their potency, as well as upon river flow and weather conditions in the delta region. The south Beaufort Sea is a sensitive area, subject to change by alterations in river properties. To prevent such changes in the sea, changes in the river must be kept minimal.

NEEDS FOR FURTHER STUDY

The extremely limited knowledge we have about Beaufort Sea biology has been mentioned several times in this report. Data used here originated mainly during a two-week period in a single summer season, and were collected only at a small number of near-shore stations. The survey should be extended into deeper water, to measure the full extent of Mackenzie influence. The full depth of water must be sampled, at least as far seaward as the edge of the shelf, to understand the oceanic influence off the delta. The time period of the study must be expanded, to include other times of the year, to follow the summer production period from its probable beginning in early summer to its probable end in early autumn. Conditions in winter, or at least near the end of winter just before summer activity begins, must be understood.

To do this the opportunity is needed to reach the Beaufort Sea at the appropriate times of the year. This means use of a proper oceanographic vessel in summer and suitable aircraft for ice landings in winter and spring. Standard oceanographic techniques are required, along with special knowledge of methods for working on the ice surface in winter.

REFERENCES

Bailey, W.B. 1957.
Oceanographic features of the Canadian archipelago. Journal of the Fisheries Research Board of Canada. 14(5): 731-769.

Barber, F.G. 1968.
On the water of Tuktoyaktuk harbour. Marine Sciences Branch, Dept. of Mines, Energy and Resources, Ottawa, Manuscript Report Series, No. 9, 32 pp.

Bursa, A. 1963.
Phytoplankton in coastal waters of the Arctic Ocean at Point Barrow, Alaska. Arctic, 16 (4): 239-262.

Cameron, W.M. 1953.
Hydrographic and oceanographic observations in the Beaufort Sea. Defence Research Board, Ottawa, Progress Report 1953: 10.

Coachman, L.K. and C.A. Barnes. 1961.
The contribution of Bering Sea water to the Arctic Ocean. Arctic, 14(3): 147-161.

Coachman, L.K. and C.A. Barnes. 1962.
Surface water of the Eurasian Basin of the Arctic Ocean. Arctic, 15(4): 251-277.

Coachman, L.K. and C.A. Barnes. 1963.
The movement of Atlantic water in the Arctic Ocean. Arctic, 16(1): 8-16.

Codispoti, L.A. and F.A. Richards. 1968.
Micronutrient distributions in the East Siberian and Laptev Seas during summer 1963. Arctic, 21(2): 67-83.

Collins, F.S. 1927.
Marine algae from Bering Strait and Arctic Ocean

collected by the Canadian Arctic Expedition, 1913-1916. Rep. Canadian Arctic Exped. 1913-18, 4(B): 16 pp.

Emery, K.O. and R.E. Stevenson. 1957.
Estuaries and lagoons. I. Physical and chemical characteristics. In: J.W. Hedgpeth (ed.) Treatise on Marine Ecology and Paleoecology. Vol. 1. Geological Society of America, Memoir, 67: 673-693.

English, T.S. 1961.
Some biological oceanographic observations in the central north Polar Sea, Drift Station Alpha, 1957-58. Arctic Institute of North America, Research Paper, 13, 80 pp.

Gudkovich, Z.M. 1955.
Results of a preliminary analysis of the deep-water hydrological observations. In: Materialy Nabliudenii Nauchno-issledovatel' Skoi Dreifuiushchei Stantsii 1950/51 Goda, (ed.) M.M. Somov, Leningrad, Ixd. 'Morskoi Transport', 1955, Vol. 1: 41-46 plus appendices pp. 48-170. (Translation by the American Meteorological Society).

Henoch, W.E.S. 1960.
Observations of Mackenzie River discharge. Canadian Geographer, 15:44-49.

Ketchum, B.H. 1967.
Phytoplankton nutrients in estuaries. In: G.H. Lauff (ed.) Estuaries. American Association for the Advancement of Science, Washington, D.C.: 329-335.

Kinney, P.J., M.E. Archelger and D.C. Burrell. 1970.
Chemical characteristics of water masses in the Amerasian Basin of the Arctic Ocean. Journal of Geophysical Research, 75: 4097-4104.

Lee, R.K.S. 1973.
General ecology of the Canadian arctic benthic marine algae. Arctic, 26(1): 32-43.

Macginitie, G.E. 1955.
Distribution and ecology of the marine invertebrates of Point Barrow, Alaska. Smithsonian Misc. Coll., 128(9): 201 pp.

Mann, A. 1925.
Marine diatoms. Rep. Canadian Arctic Exped. 1913-18, 4(F): 33 pp.

Reeder, S.W., B. Hitchon and A.A. Levison. 1972.
Hydrogeochemistry of the surface waters of the Mackenzie River drainage basin, Canada - 1. Factors controlling inorganic composition. Geochimica et Cosmochimica Acta, 36: 825-865.

Riley, G.A. 1937. The significance of the Mississippi River drainage for biological conditions in the northern Gulf of Mexico. Journal of Marine Research, 1: 60-74.

Shih, C.-T, A.J.G. Figueira and E.H. Grainger. 1971.
A synopsis of Canadian marine zooplankton. Fish. Res. Bd. Canada, Bull., 176: 264 pp.

Strickland, J.D.H. and T.R. Parsons. 1968.
A practical handbook of seawater analysis. Fisheries Research Board of Canada, Bulletin, 167, 311 pp.

Tully, J.P. 1952.
Oceanographic data of the western Canadian arctic region, 1935-37. Journal of the Fisheries Research Board of Canada, 8(5): 378-382.

U.S. Navy Hydrographic Office. 1954.
Oceanographic observations, U.S.S. Burton Island, 1950-1953. U.S. Navy Hydrographic Office Publication. 618-C, 309 pp.

TABLE 1

Physical and Chemical Data Collected
in the South Beaufort Sea
in July 1973

Table 1

Physical and Chemical Data Collected in the South Beaufort Sea in July of 1973

Station: 526 Date: 20/7 Time: 1640 GMT

Station depth: 8m Air temp.: 4.5°C Secchi: 0.3m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	11.2	2.8	6.8	91	-
3	11.3	2.8	7.1	93	-
5	11.2	3.0	7.1	93	-
7	4.6	16.5	7.1	87	-

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. a (mg/m^3)	POC (mg/m^3)
0	0.00	0.6	34.6	2.05	-
3	0.00	0.6	34.4	2.22	-
5	0.00	0.1	11.4	2.36	-
7	0.00	0.8	-	1.64	-

Table 1 (cont'd)

Station: 527 Date: 20/7 Time: 2115 GMT

Station depth: 5m Air temp.: 10.0°C Secchi: 0.3m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	11.6	1.5	6.7	88	0.32
3	11.5	1.7	7.7	101	1.47
5	10.0	5.0	7.7	100	0.25

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. *a* (mg/m^3)	POC (mg/m^3)
0	0.00	2.4	30.4	2.49	514
3	0.00	2.7	21.4	2.28	293
5	0.00	2.1	16.9	2.15	503

Table 1 (cont'd)

Station: 528 Date: 22/7 Time: 2125 GMT

Station depth: 7m Air temp.: 6.7°C Secchi: 0.3m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	9.0	5.8	7.8	99	0.21
3	8.1	6.6	7.8	98	0.74
5	8.0	6.8	7.9	99	0.59
7	8.0	6.9	8.1	101	0.63

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. *a* (mg/m^3)	POC (mg/m^3)
0	0.00	1.4	23.2	2.93	220
3	0.00	0.7	7.2	1.61	404
5	0.00	1.3	15.4	1.43	409
7	0.00	1.3	15.4	2.01	315

Table 1 (cont'd)

Station: 529 Date: 23/7 Time: 0130 GMT

Station depth: 12m Air temp.: 5.9°C Secchi: 0.3m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	7.7	9.0	7.9	99	0.21
1	7.7	9.0	-	-	-
3	7.5	9.2	7.9	99	0.36
5	7.5	9.3	8.1	102	0.15
7	7.5	9.5	-	-	-
10	7.0	9.6	8.0	99	0.27

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. a (mg/m^3)	POC (mg/m^3)
0	0.00	0.1	19.2	1.40	221
1	-	-	-	-	-
3	0.00	0.0	18.8	1.39	278
5	0.00	0.0	19.3	1.80	185
7	-	-	-	-	-
10	0.00	0.0	19.5	1.24	191

Table 1 (cont'd)

Station: 530 Date: 23/7 Time: 1540 GMT

Station depth: 9m Air temp.: 7.0°C Secchi: 0.7m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	7.6	8.8	8.0	100	0.19
1	7.5	8.8	-	-	-
3	6.9	10.6	8.1	101	0.06
5	6.8	11.2	8.2	103	0.13
7	6.8	11.8	-	-	-
9	6.6	12.7	8.3	104	0.25

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. a (mg/m^3)	POC (mg/m^3)
0	0.00	0.1	16.2	1.76	82
1	-	-	-	-	-
3	0.00	0.1	22.4	1.03	125
5	0.00	0.0	20.7	0.97	127
7	-	-	-	-	-
9	0.00	0.0	17.7	0.45	125

Table 1 (cont'd)

Station: 531 Date: 23/7 Time: 1950 GMT

Station depth: 15m Air temp.: 9.5°C Secchi: 1.0m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	7.0	12.8	8.2	104	0.27
1	6.8	13.8	-	-	-
3	6.6	14.0	-	-	-
5	6.5	14.1	8.2	104	0.13
10	6.3	14.5	8.3	105	0.13
14	6.3	15.5	8.2	104	0.17

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. a (mg/m^3)	POC (mg/m^3)
0	0.00	0.0	20.6	-	119
1	-	-	-	-	-
3	-	-	-	-	-
5	0.00	0.0	20.2	0.56	127
10	0.01	0.0	20.0	0.57	159
14	0.00	0.0	19.9	0.91	131

Table 1 (cont'd)

Station: 532 Date: 24/7 Time: 0045 GMT

Station depth: 36m Air temp.: 10.6°C Secchi: 14m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	5.0	25.8	8.5	113	0.42
1	5.0	26.8	-	-	-
3	5.0	27.0	-	-	-
5	5.2	27.3	8.3	111	0.40
7	5.3	27.6	-	-	-
10	3.6	28.3	9.0	117	0.57
15	2.9	31.5	-	-	-
20	0.4	32.2	9.8	121	0.78
30	-0.4	32.4	-	-	-
35	-0.5	32.4	8.8	106	1.43

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. *a* (mg/m^3)	POC (mg/m^3)
0	0.02	0.0	7.6	0.45	121
1	-	-	-	-	-
3	-	-	-	-	-
5	0.01	0.0	6.5	0.33	171
7	-	-	-	-	-
10	0.03	0.0	6.6	-	129
15	-	-	-	-	-
20	0.08	0.3	8.7	0.41	211
30	-	-	-	-	-
35	0.12	2.4	14.6	0.57	211

Table 1 (cont'd)

Station: 533 Date: 24/7 Time: 0420 GMT

Station depth: 42m Air temp.: 8.5°C Secchi: 13.5m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	6.2	26.7	8.3	114	0.42
3	6.1	26.7	-	-	-
5	5.8	26.9	8.3	117	0.42
7	5.7	26.9	-	-	-
10	5.0	27.8	8.5	113	0.44
15	2.2	31.8	-	-	-
20	1.6	31.9	9.9	125	0.46
30	-0.8	32.7	8.8	105	1.26
40	-0.8	32.8	8.5	102	1.26

Depth (m)	NO_2-N) (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. a (mg/m^3)	POC (mg/m^3)
0	0.00	0.0	7.6	0.00	128
3	-	-	-	-	-
5	0.00	0.0	7.3	0.32	142
7	-	-	-	-	-
10	0.00	0.0	6.3	0.15	138
15	-	-	-	-	-
20	0.00	0.0	2.8	0.12	-
30	0.14	3.1	15.5	-	229
40	0.08	2.7	14.3	-	210

Table 1 (cont'd)

Station: 534 Date: 24/7 Time: 2315 GMT

Station depth: 7m Air temp.: 17.8°C Secchi:

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	11.9	1.7	7.4	98	0.76
1	11.8	1.7	-	-	-
3	11.2	1.7	7.5	99	0.50
5	5.5	9.2	7.8	93	0.17
7	5.4	9.3	-	-	-

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. *a* (mg/m^3)	POC (mg/m^3)
0	0.00	2.4	33.8	1.53	142
1	-	-	-	-	-
3	0.00	2.5	33.0	0.87	171
5	0.00	1.5	38.8	1.40	106
7	-	-	-	-	-

Table 1 (cont'd)

Station: 535 Date: 25/7 Time: 2330 GMT

Station depth: 6m Air temp.: 22.0°C Secchi:

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	14.0	1.2	6.9	96	1.05
1	13.6	1.2	-	-	-
3	8.0	3.2	7.5	92	0.15
5	6.3	7.8	7.4	89	0.23

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. *a* (mg/m^3)	POC (mg/m^3)
0	0.00	3.1	26.1	2.25	278
1	-	-	-	-	-
3	0.00	1.1	16.6	1.35	163
5	0.00	0.5	16.3	1.80	194

Table 1 (cont'd)

Station: 536 Date: 26/7 Time: 0300 GMT

Station depth: 9m Air temp.: 19.0°C Secchi: 0.3m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	14.0	1.7	7.4	103	0.65
1	10.9	1.9	-	-	-
3	8.5	4.5	7.9	98	0.55
5	5.4	8.0	8.1	97	0.00
8	2.8	10.8	8.3	93	0.21

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. a (mg/m^3)	POC (mg/m^3)
0	0.00	2.9	32.3	1.19	-
1	-	-	-	-	-
3	0.00	2.2	30.0	0.56	182
5	0.00	0.7	23.1	1.77	138
8	0.00	0.4	21.6	1.59	184

Table 1 (cont'd)

Station: 537 Date: 26/7 Time: 0615 GMT

Station depth: 9m Air temp.: 16.0°C Secchi: 0.3m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	11.4	1.2	7.4	97	-
1	11.2	1.2	-	-	-
3	9.8	1.8	7.7	97	0.00
5	4.9	8.0	8.1	95	0.00
7	4.7	8.6	-	-	-
9	0.6	24.4	8.5	99	0.32

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. a (mg/m^3)	POC (mg/m^3)
0	0.00	2.0	11.6	1.66	353
1	-	-	-	-	-
3	0.00	2.5	-	1.40	213
5	0.00	0.6	12.4	0.70	110
7	-	-	-	-	-
9	0.02	2.3	15.8	0.96	127

Table 1 (cont'd)

Station: 538 Date: 26/7 Time: 1905 GMT

Station depth: 5m Air temp.: 21.0°C Secchi: 0.2m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	15.6	0.1	6.9	99	0.32
1	14.7	0.1	-	-	-
3	13.2	0.2	7.4	100	0.84
5	8.5	4.6	7.3	91	0.53

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. a (mg/m^3)	POC (mg/m^3)
0	0.00	2.4	23.4	1.67	-
1	-	-	-	-	-
3	0.00	3.6	26.4	-	-
5	0.00	3.6	24.4	-	-

Table 1 (cont'd)

Station: 539 Date: 26/7 Time: 2240 GMT

Station depth: 3m Air temp.: 23.0°C Secchi: 0.2m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	17.2	<0.1	7.1	105	1.49
1	17.1	<0.1	-	-	-
3	16.6	<0.1	7.1	104	0.84

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. a (mg/m^3)	POC (mg/m^3)
0	0.00	3.7	27.9	2.07	-
1	-	-	-	-	-
3	0.00	3.6	24.3	2.30	421

Table 1 (cont'd)

Station: 540 Date: 27/7 Time: 0215 GMT

Station depth: 4m Air temp.: 22.0°C Secchi: 0.2m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	16.4	0.1	7.5	110	0.63
1	16.3	0.1	-	-	-
3	14.6	0.1	7.6	106	0.38
4	8.7	10.5	7.7	99	0.48

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. a (mg/m^3)	POC (mg/m^3)
0	0.02	3.5	33.2	1.71	-
1	-	-	-	-	-
3	0.00	3.7	31.6	1.55	-
4	0.00	2.5	24.2	2.18	-

Table 1 (cont'd)

Station: 541 Date: 27/7 Time: 1840 GMT

Station depth: 34m Air temp.: 12.0°C Secchi: 1.0m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	10.0	6.8	8.3	109	0.00
3	7.0	9.0	-	-	-
5	3.5	13.4	8.6	101	0.17
7	1.9	18.2	-	-	-
10	0.4	25.2	9.9	115	0.57
15	0.0	28.8	-	-	-
20	-0.1	30.0	9.9	119	0.84
30	-0.1	30.1	9.9	118	0.61

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. a (mg/m^3)	POC (mg/m^3)
0	0.00	0.3	32.8	3.24	-
3	-	-	-	-	-
5	0.00	0.6	30.2	1.19	-
7	-	-	-	-	-
10	0.00	1.3	13.4	0.22	-
15	-	-	-	-	-
20	0.06	3.4	11.9	0.05	-
30	0.06	3.6	11.6	-	-

Table 1 (cont'd)

Station: 542 Date: 27/7 Time: 2310 GMT

Station depth: 94m Air temp.: 11.0°C Secchi: 2.2m

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	7.1	12.0	9.3	117	0.13
3	7.0	12.0	-	-	-
5	5.0	14.3	9.3	113	0.13
7	1.4	23.1	-	-	-
10	-0.2	29.5	10.6	127	0.61
20	-0.2	30.5	10.5	127	0.65
30	-0.1	30.9	10.5	127	1.09
50	0.1	31.1	10.0	120	0.90
75	-1.25	31.456	9.7	114	1.39
90	-1.22	32.335	9.4	111	-

Depth (m)	NO_2-N (µg-at/l)	NO_3-N (µg-at/l)	Si (µg-at/l)	Ch. a (mg/m^3)	POC (mg/m^3)
0	0.00	0.0	17.7	0.59	162
3	-	-	-	-	-
5	0.00	0.1	19.1	0.81	-
7	-	-	-	-	-
10	0.05	2.1	12.1	0.16	-
20	0.06	2.1	9.5	0.00	-
30	0.08	2.4	14.0	-	-
50	0.09	6.8	19.0	-	-
75	0.09	-	15.2	-	-
90	-	9.5	24.3	-	-

Table 1 (cont'd)

Station: 543A Date: 23/7 Time: 2300 GMT

Station depth: - Air temp.: 10.0°C Secchi: -

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	7.9	18.9	-	-	-
3	6.2	25.7	-	-	-
5	5.3	27.1	-	-	-
10	4.8	27.6	-	-	-
15	-0.2	31.8	-	-	-

Table 1 (cont'd)

Station: 543B Date: 23/7 Time: 2320 GMT

Station depth: - Air temp.: 10.0°C Secchi: -

Depth (m)	Temp. (°C)	Salinity (°/oo)	Oxygen (ml/l)	Oxygen (%)	PO_4-P (µg-at/l)
0	7.0	23.0	-	-	-
3	5.8	26.8	-	-	-
5	5.2	27.4	-	-	-
10	4.9	27.6	-	-	-
15	0.0	31.7	-	-	-

TABLE 2

Zooplankton Species Collected in
the South Beaufort Sea in July 1973

Table 2

Species	Depth (m)	Numbers per m^3, at stations 526	527	528	529	530	531	532
		5-0	4-0	6-0	10-0	8-0	14-0	34-0
Hydrozoa								
Sarsia princeps								+
Halitholus cirratus						2		
Euphysa flammea								
Obelia sp.								4
Aglantha digitale								9
Aeginopsis laurenti								1
Dimophyes arctica								
Mollusca								
Spiratella helicina								4
Annelida								
Polychaete larvae						4	8	96
Branchiopoda								
Daphnia sp.								
Copepoda								
Acartia clausi				23	1	4	2	
A. longiremis							2	
Derjuginia tolli							1	
Calanus glacialis								16
C. hyperboreus								13
Limnocalanus macrurus		56	1091	4484	140	117	33	
Diaptomus sp.								
Euchaeta glacialis								
Metridia longa								8
Microcalanus pygmaeus								
Pseudocalanus minutus					6	10	52	283
Epischura sp.								
Eurytemora herdmani		5803	426		27	155	10	
Ectinosoma sp.								
Cyclops sp.		20226	845	23	10		1	
Cyclopina sp.						4		
Oithona similis							1	189
Oncaea borealis								41
Copepod nauplii		23493	599	47	79	963	714	2217

Table 2 (cont'd)

	Numbers per m^3, at stations						
	526	527	528	529	530	531	532
Species Depth (m)	5-0	4-0	6-0	10-0	8-0	14-0	34-0
Cirripedia							
Balanus sp.							13
Mysidacea							
Mysis sp.		8	14				
Cumacea							
Diastylis sp.		11	2				2
Amphipoda							
Pseudalibrotus glacialis							
Monoclodes sp.		49	2			4	
Hyperia galba							+
H. medusarum							
Parathemisto abyssorum							
P. Libellula							
Euphausiacea							
Thysanoessa raschi							
Decapoda							
Sabinea sp.							
Chaetognatha							
Eukrohnia hamata							
Sagitta elegans							+
Copelata							
Fritillaria borealis							
Oikopleura vanhoeffeni							38
tunicate larvae							

Table 2 (cont'd)

Species	Depth (m)	Numbers per m^3, at stations 533	534	535	536	537	538	540
		40-0	6-0	5-0	8-0	8-0	5-0	4-0
Hydrozoa								
Sarsia princeps								
Halitholus cirratus								
Euphysa flammea								
Obelia sp.								
Aglantha digitale		5						
Aeginopsis laurenti								
Dimophyes arctica								4
Mollusca								
Spiratella helicina								
Annelida								
polychaete larvae		7						
Branchiopoda								
Daphnia sp.							55	64
Copepoda								
Acartia clausi								
A. longiremis								
Derjuginia tolli								
Calanus glacialis		14						
C. hyperboreus		13						
Limnocalanus macrurus			825	736	655	1186	12	8
Diaptomus sp.							+	
Euchaeta glacialis								
Metridia longa		1						
Microcalanus pygmaeus								
Pseudocalanus minutus		22		28				
Epischura sp.							6	16
Eurytemora herdmani							6	100
Ectinosoma sp.		4						
Cyclops sp.			24	28		54	345	321
Cyclopina sp.								
Oithona similis		131						
Oncaea borealis		100						
copepod nauplii		1105	118			35	229	1021

Table 2 (Cont'd)

Species	Depth (m)	Numbers per m^3, at stations						
		533	534	535	536	537	538	540
		40-0	6-0	5-0	8-0	8-0	5-0	4-0
Cirripedia								
Balanus sp.								
Mysidacea								
Mysis sp.			9	3	5	18	6	4
Cumacca								
Diastylis sp.								
Amphipoda								
Pseudalibrotus glacialis								
Monoculodes sp.				3				
Hyperia galba								
H. medusarum								
Parathemisto abyssorum								
P. libellula								
Euphausiacea								
Thysanoessa raschi								
Decapoda								
Sabinea sp.								
Chaetognatha								
Eukrohnia hamata								
Sagitta elegans								
Copelata								
Fritillaria borealis								
Oikopleura vanhoeffeni								
tunicate larvae								

Table 2 (Cont'd)

	Numbers per m^3, at stations					
	541	541	542	542	542	542
Species Depth (m)	30-10	10-0	90-75	75-50	50-10	10-0
Hydrozoa						
Sarsia princeps	1					
Halitholus cirratus						
Euphysa flammea				6		
Obelia sp.						
Aglantha digitale	1				+	
Aeginopsis laurenti	+				+	
Dimophyes arctica	+				+	
Mollusca						
Spiratella helicina	9		+			
Annelida						
polychaete larvae	28	28			35	85
Branchiopoda						
Daphnia sp.						
Copepoda						
Acartia clausi						
A. longiremis						
Derjuginia tolli	37	14			6	
Calanus glacialis	12		14			
C. hyperboreus	17	42		143	34	56
Limnocalanus macrurus		693				692
Diaptomus sp.						
Euchaeta glacialis				10	1	
Metridia longa	6		4	24	4	
Microcalanus pygmaeus	98	14	79	135	181	28
Pseudocalanus minutus	194			34	19	155
Epischura sp.						
Eurytemora herdmani		14				14
Ectinosoma sp.		14	75	27	7	28
Cyclops sp.		38				
Cyclopina sp.		14	7			
Oithona similis	411	183		87	282	28
Oncaea borealis	239	42		182	228	14
copepod nauplii	6875	6393			1453	8600

Table 2 (Cont'd)

Species	Depth (m)	Numbers per m^3, at stations 541 30-10	541 10-0	542 90-75	542 75-50	542 50-10	542 10-0
Cirripedia							
Balanus sp.							
Mysidacea							
Mysis sp.			1				+
Cumacca							
Diastylis sp.							
Amphipoda							
Pseudalibrotus glacialis				+			
Monoculodes sp.							
Hyperia galba				+			
H. medusarum						+	
Parathemisto abyssorum				+			
P. libellula				+			
Euphausiacea							
Thysanoessa raschi		1					
Decopoda							
Sabinea sp.				+			
Chaetognatha							
Eukrohnia hamata				+			
Sagitta elegans				+			
Copelata							
Fritillaria borealis				56	40	32	14
Oikopleura vanhoeffeni		1		13		6	
tunicate larvae				8	6		

Part IIIb

Distribution and Abundance of Heterotrophic Bacteria in the South Beaufort Sea

by

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TABLE OF CONTENTS

LIST OF FIGURES

SUMMARY

Microbiological samplings were conducted during a cruise on the South Beaufort Sea in July, 1973. Total viable counts on marine and freshwater cultivation media were made to provide an estimate of the distribution and abundance of heterotrophic bacteria in the waters sampled. A large biomass of freshwater bacteria was found to be discharged into the Beaufort Sea from the Mackenzie River, but this biomass did not appear to persist in the marine waters. A relatively uniform abundance of marine bacteria was found in the areas sampled. Preliminary studies suggest that this flora is similar to the flora observed in the Eskimo Lakes.

INTRODUCTION

A microbiological sampling of the South Beaufort Sea was conducted during the last part of July, 1973, as part of the oceanographic cruise described in Part a of this report, on the vessel, "North Star of Herschel Island". At designated stations, where plankton collections were made and water samples taken for chemical and physical data, water from similar depths in the water columns was processed for microbiological examination. The objectives of this aspect of the cruise were to:

1. obtain estimates of the viable heterotrophic populations of bacteria in the South Beaufort Sea;

2. determine the influence of the freshwater flow of the Mackenzie River into the South Beaufort Sea on the distribution of heterotrophic populations of bacteria;

3. compare microbial populations of the South Beaufort Sea with more intensively studied populations in the Eskimo Lakes, an inlet of the South Beaufort Sea.

Bacterial heterotrophs and fungi are the only known forms of organisms involved in the biological degradation of petroleum and petroleum products in natural waters. A study of bacterial distribution was a prerequisite to assessing the fate of residual petroleum in the South Beaufort Sea.

CURRENT STATE OF KNOWLEDGE

Prior to the cruise undertaken this summer, no information concerning the microbial flora of the South Beaufort Sea was available. Kriss (1963) described the quantitative distribution of heterotrophic bacteria in the North Beaufort Sea from spatial and seasonal samplings conducted from floating ice stations in 1955-56. Although a limited number of bacteria were found in water columns during July and September, no bacteria were recovered in the months of April or May and led Kriss to conclude that microbial activity occurred

only during the period of intense light in the summer months. The techniques employed to obtain this information, however, did not discern the psychrophilic, or cold-loving, populations of cells in these waters and the described data does not represent a true estimate of the microbial flora. More recently, a report by Robertson et al. (1973) of samplings in the waters adjacent to Point Barrow, Alaska, indicated the existence of a heterotrophic flora, but at a low level of activity. In view of the limited knowledge of the microbiology of the Beaufort Sea, the present study was initiated.

METHODS AND SOURCES OF DATA

1. Plan of cruise

The details of the cruise schedule, together with methods employed to determine physical and chemical parameters of water samples, are presented in Part a of this report.

2. Culture media

To enhance the multiplication and colony-formation of marine bacteria, ZoBell Marine Broth 2216E (Difco), containing a formulation of seawater salts, was prepared with deionized water as an agar medium. A freshwater or terrestrial cultivation medium consisting of Plate Count Agar (Difco) was also prepared with deionized water. Both media were dispensed in Petri plates (100 mm dia.) at microbiological facilities of the Arctic Biological Station in Inuvik, N.W.T. Prepared plates were taken on board the cruise vessel at Inuvik and stored at 5.0°C until required.

3. Sampling procedure

During the occupation of a station, water samples were taken aseptically with Niskin SS 1.5 sterile bag-samplers at various depths of the water column prior to disturbances of the water column by other oceanographic samplings. These samples were kept chilled and processed within one half hour of removal from the water column. A spin-plate technique was employed to dispense 0.1 ml of water sample with a

cold 0.2 ml pipette on the surface of a cold agar plate which was then placed in a 5.0°C refrigerator. Upon absorption of the aliquot of water by the agar medium, the plate was inverted and incubation was continued at 5.0°C. Quadruplicate spin-plates were made of each sample on both plating media. At the termination of the cruise, plates were transferred in a chilled condition to the laboratory in Inuvik and incubation was continued to completion at three weeks. At that time, the plates were examined and those with an uneven distribution of colonies were discarded. The colonies of three plates of a replicate set were enumerated, averaged, and the mean value was expressed as the log number of colony-forming units (C.F.U.) per one litre of water sample.

RESULTS

In the past experience of this laboratory, the plating media employed in this study have been the most successful media in the cultivation of heterotrophic organisms from estuarine and marine waters. Although the counts obtained may not represent the total heterotrophic population, but rather those colony-forming units i.e. cells or clumps of cells, capable of colony formation on the media employed, no other media have been found by this laboratory to yield a higher count. Indigenous marine bacteria were defined in this study as cells which required seawater for their cultivation. This population of heterotrophic bacteria was represented by the counts obtained on ZoBell Marine Medium.

In the absence of a salt water formulation, the colonies cultivated on Plate Count Agar (PCA) were considered to be representative of the freshwater or terrestrial flora of bacteria present in the estuarine areas sampled.

The counts obtained on ZoBell Marine Medium and PCA were compared and related to the salinity values obtained at various depths of the stations occupied (see Part a of this report). The results obtained in four transects of the cruise are seen in Figures 1 to 4. Mean values and standard deviations of plate counts at each station are presented in the appendix.

Stations 527 to 531, (Figure 1) represent a transect parallel to the Tuktoyaktuk Peninsula and the discharge of the East Channel of the Mackenzie River into the Beaufort Sea. Increasing salinity values corresponded to decreasing counts on PCA as the freshwater influence of the Mackenzie channel diminished. The high counts obtained on ZoBell plates appeared to demonstrate the presence of a marine flora in the mixed marine and freshwater in this region, but the growth of some freshwater flora on these plates was not discounted. In the transect of Stations 536-540 across the discharge of the West Channel of the Mackenzie River into Mackenzie Bay, (Fig. 2), the salinity values obtained again corresponded to a pattern of counts on the two plating media. The depressed counts on ZoBell plates from essentially freshwater samplings at Stations 538, 539 and 540 were in contrast to the higher counts obtained on PCA and the difference in count indicated a population of freshwater microorganisms which were unable to multiply on the marine medium.

In Figures 3 and 4, the results of two transects from the coast into the deeper marine waters of the Beaufort Sea are demonstrated. With increasing salinity of the water, and decreasing temperatures (see data in Part a), the inability of freshwater microorganisms to persist and multiply in the marine environment of the Beaufort Sea was suggested by the paucity of counts obtained on PCA plates prepared from these waters. At the same time, the flora cultivated on ZoBell medium represented indigenous marine bacteria of the Beaufort Sea.

DISCUSSION

The results of this study have established a total viable count of heterotrophic bacteria in the waters of the South Beaufort Sea during July. Although sampling techniques and cultivation media vary among investigations, the abundance of heterotrophs is comparable to values obtained by investigations of estuaries and offshore areas along the New England coast (Kaneko and Colwell 1973; Sieburth, 1967; Atlas and Bartha, 1972). This does not imply, however, that similar varieties of heterotrophs or similar rates of activity by these cells can be demonstrated. Experiments in this

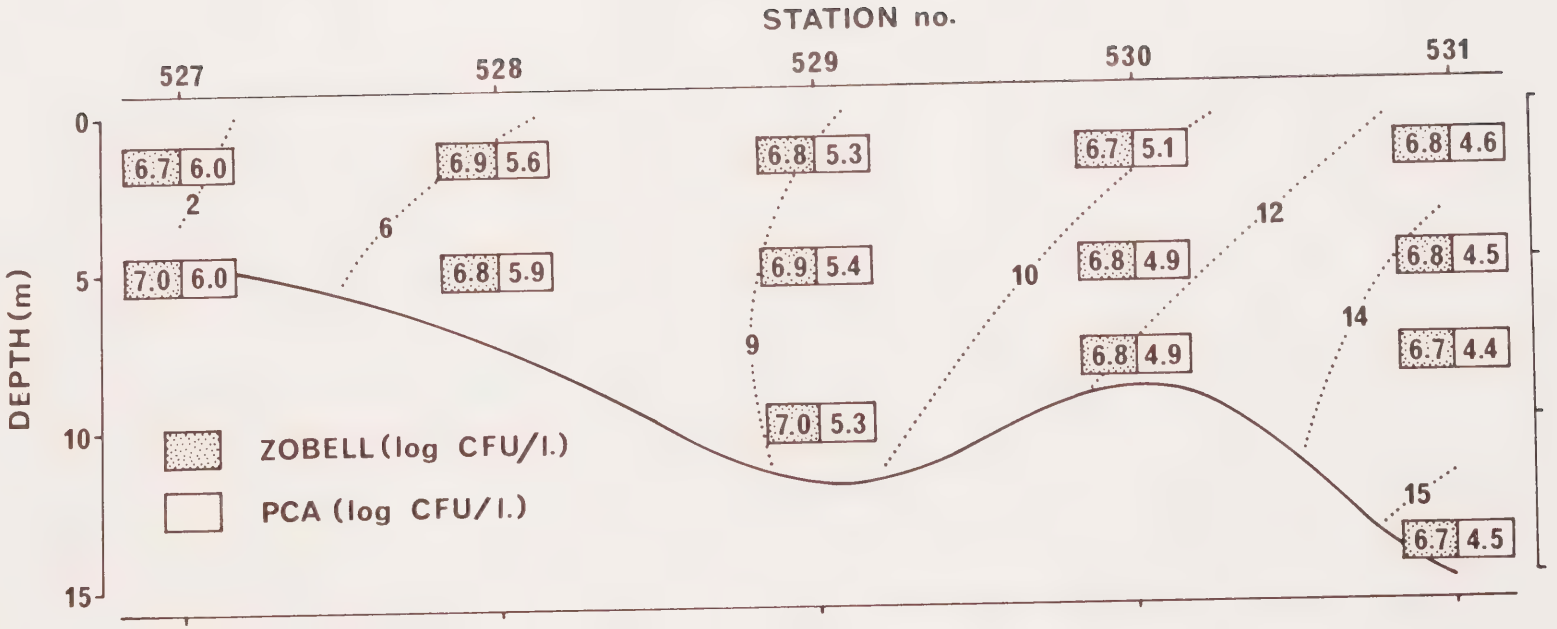

Figure 1. Salinity profile and total viable bacterial counts of water between Stations 527 and 531. (See map in Part A.) Salinity values (°/₀₀, dotted lines) are interpolated between stations. Colony counts obtained on ZoBell and PCA media at various depths are expressed as the log number of colony-forming units (C.F.U.) per one litre of sampled water. Solid line indicates approximate depth to bottom sediment. Salinity values were obtained from Dr. E. H. Grainger.

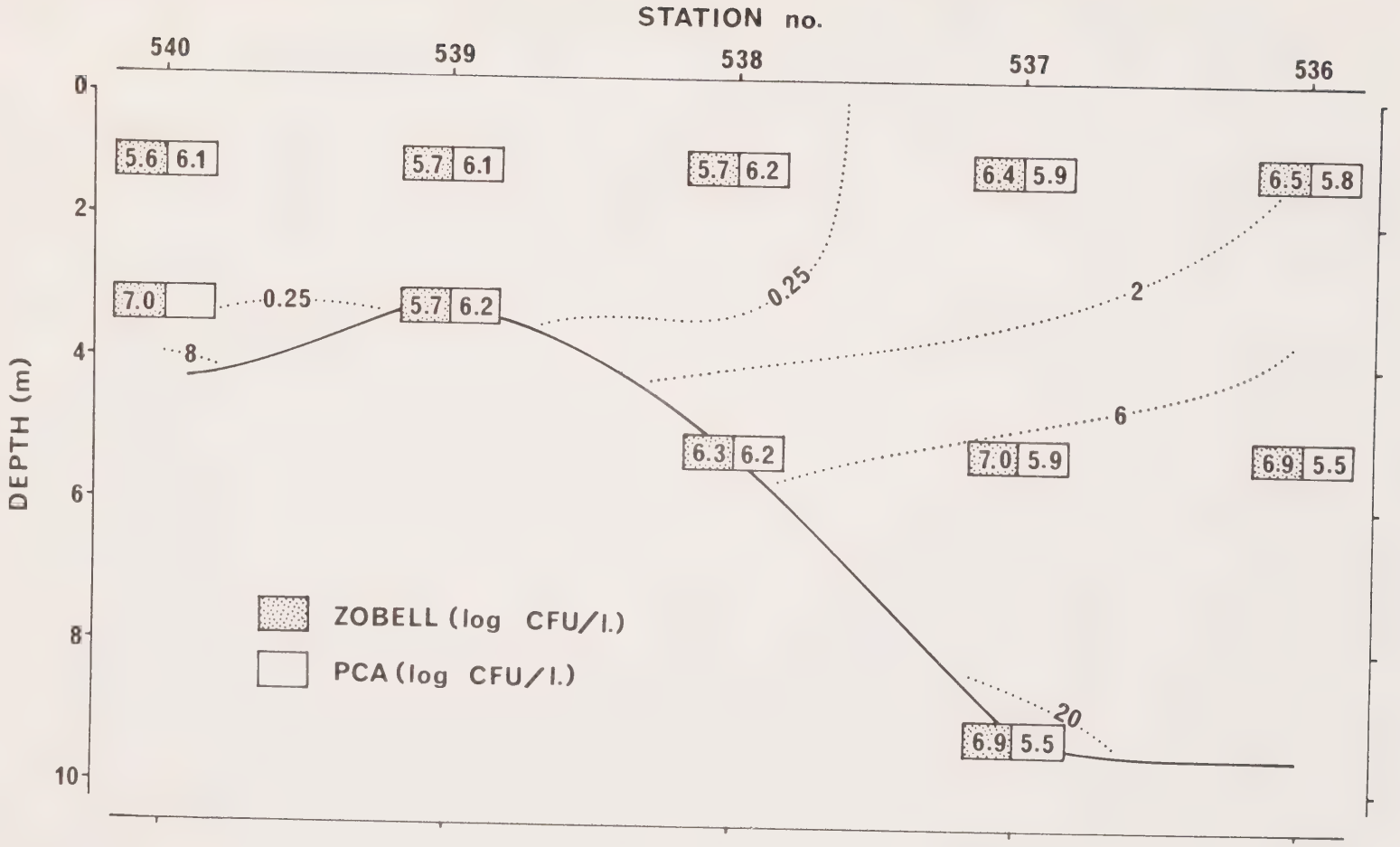

Figure 2. Salinity profile and total viable bacterial counts of water between Stations 540 and 536. See legend to Figure 1.

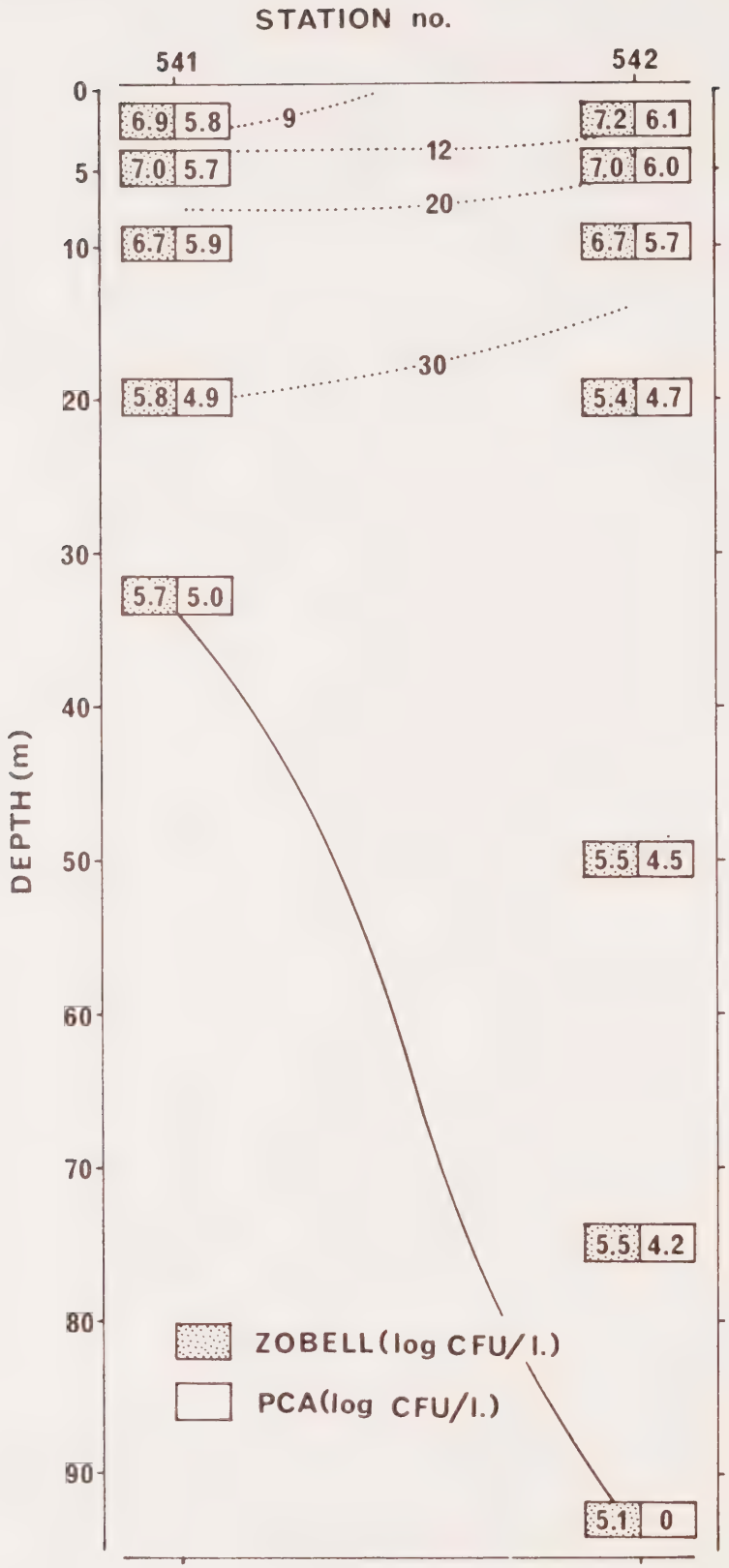

Figure 3. Salinity profile and total viable bacterial counts of water between Stations 541 and 542. See legend to Figure 1.

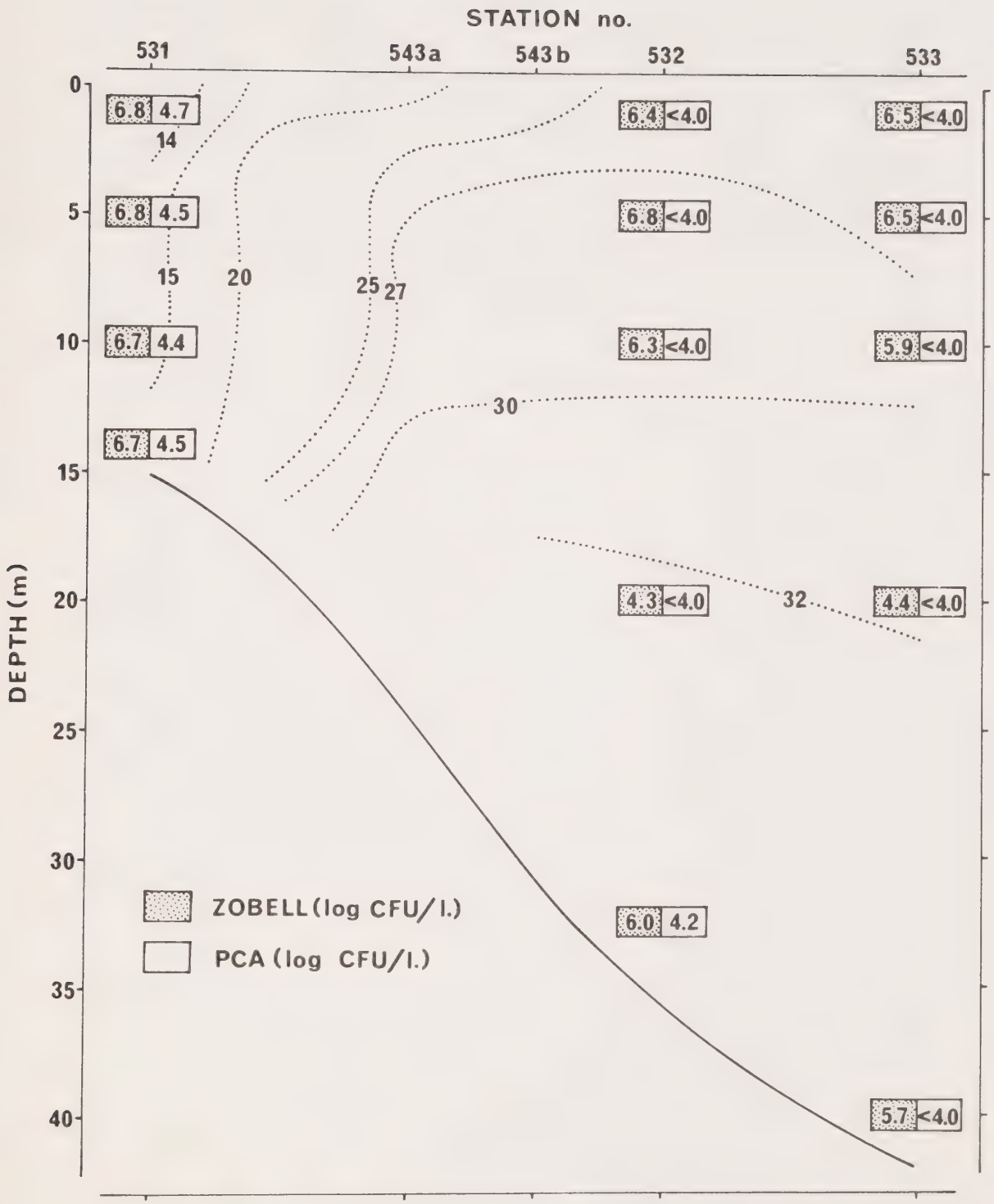

Figure 4. Salinity profile and total viable bacterial counts of water between Stations 531 and 533. See legend to Figure 1.

laboratory suggest that a majority of heterotrophs isolated from Arctic marine waters are obligately psychrophilic in all seasons (data not reported), a situation not found in warmer latitudes.

The data suggest that the discharge of the Mackenzie River into the Beaufort indeed contains a large biomass of freshwater heterotrophs. These cells, however, were not observed to persist in marine waters. Similar observations in estuarine environments have been reported by Jones (1971). The counts of marine flora were found to be high in the areas of mixed water and may have contained freshwater flora. However, the river discharge was not found to substantially alter the counts of marine flora in the areas sampled except at Stations 537 to 540 where the lowest salinity values were recorded.

Counts of marine flora from all stations except 537 to 540 were characterized by the predominant growth of an orange-pigmented colony on the plating medium. Similar levels of abundance of this colony type on ZoBell marine medium were observed during July, 1973, from samples obtained in stations on the Eskimo Lakes. This type of colony was never observed on plates of PCA and work from this laboratory has shown this bacterial species to be truly marine in its cultural requirements. An assessment of the metabolic characteristics of this strain and other heterotrophic isolates from the Beaufort Sea and Eskimo Lakes is presently being conducted, but preliminary data would suggest that a relatively homogenous marine flora exists in all regions sampled.

Studies in the Eskimo Lakes in 1973 have demonstrated a heterotrophic population capable of degrading crude petroleum (see other report in this series by Bunch and Harland). It seems probable that a similar flora exists in the South Beaufort Sea. Off-shore oil spills or petroleum and petroleum products discharged from the Mackenzie would probably be subjected to a degree of degradation by an indigenous marine flora at a rate yet to be determined. The freshwater flora in the estuary would probably not participate in an oil-degrading or oleoclastic process due to various factors in the estuarine environment which would tend to inhibit growth and multiplication.

CONCLUSIONS

1. A marine heterotrophic population of bacteria has been established to exist at a relatively uniform level of abundance in the sampled areas of the South Beaufort Sea.

2. The Mackenzie River contributes a large biomass of freshwater heterotrophs to the estuarine system but this population probably does not persist.

3. The heterotrophic flora of the South Beaufort Sea appears to approximate that found in the Eskimo Lakes and an oleoclastic potential probably exists. This conclusion, however, is tentative and subject to the completion of the study.

IMPLICATIONS AND RECOMMENDATIONS

Studies undertaken in various marine environments have demonstrated the ability of indigenous bacteria to degrade crude oil to varying degrees. Attempts have been made to extrapolate these studies to conditions in the Arctic Ocean (Atlas and Bartha, 1972), but such extrapolations are subject to doubt. This study was undertaken to determine if the distribution and abundance of heterotrophic bacteria in the South Beaufort Sea was sufficient to warrant further investigation of the oleoclastic potential of these organisms and to ascertain those areas where future investigations could best be accomplished. The results of this and other studies in this laboratory tend to suggest the possibility of biological degradation of petroleum in Arctic marine waters but further studies are required. The persistence of petroleum in the ecosystem of the Beaufort Sea cannot be ascertained at this time. However, some microbial modification of weathered, residual petroleum might be expected.

NEEDS FOR FURTHER STUDY

To increase knowledge of biological degradation of petroleum in the Beaufort ecosystem, further studies

are required to:

1. determine seasonal fluctuations of the biomass of heterotrophic bacteria;

2. attempt to quantitate the abundance of oleoclastic heterotrophs;

3. determine the minimum and optimum temperatures for biological degradation by various heterotrophic isolates;

4. devise a procedure for measuring the in situ rate of biological degradation of petroleum.

These studies could best be accomplished by two or more cruises similar to the one undertaken last year. The data obtained would be supplemented by additional laboratory work.

REFERENCES

Atlas, R.M. and R. Bartha. 1972.
Biodegradation of petroleum in seawater at low temperatures. Can. J. Microbiol. 18:1851-1855.

Jones, G.E. 1971.
The fate of freshwater bacteria in the sea. In: Developments in Industrial Microbiology. 12:141-151.

Kaneko, T. and R.R. Colwell. 1973.
Ecology of Vibrio parahaemolyticus in Chesapeake Bay. Journ. Bacteriol. 113: 24-32.

Kriss, A.E. 1963.
Marine Microbiology. pp. 31-34. Oliver and Boyd, London, G.B.

Robertson, B., S. Arhelger, P.J. Kinney, and D.K. Button. 1973.
Hydrocarbon biodegradation in Alaskan waters. In: The Microbial Degradation of Oil Pollutants. pp. 171-184. D.G. Ahearn and S.P. Meyer (eds.). Center for Wetland Resources. Publication No. LSU-SG-73-01. Louisiana State U. Baton Rouge, La.

Sieburth, J. McN. 1967.
Seasonal selection of estuarine bacteria by water temperature. J. exp. mar. Biol. Ecol. 1: 98-121.

APPENDIX

Plate counts obtained from 0.1 ml aliquots of water samples taken from all stations occupied in the South Beaufort Sea during July, 1973. Inoculated plates of ZoBell Marine Medium 2216E and PCA were incubated at 5.0°C for three weeks. The mean of the counts of three replicate plates is given in each case, together with the standard deviation and the common logarithm of the mean multiplied by 10^4.

Station Number	Depth (m)	ZoBell			PCA		
		Mean	S.D.	$Log_{10} x10^4$	Mean	S.D.	$Log_{10} x10^4$
526	1	80.0	-	5.90	68.3	8.5	5.84
	5	126.0	-	6.10	40.3	8.7	5.61
527	1	461.3	54.5	6.66	104.3	14.6	6.02
	5	1037.0	-	7.02	102.3	4.5	6.01
528	1	813.3	108.0	6.91	41.7	4.0	5.62
	5	704.0	-	6.85	71.0	5.0	5.85
529	1	664.7	56.7	6.82	18.3	1.5	5.26
	5	803.3	48.0	6.90	25.3	2.9	5.40
	10	894.3	59.2	6.95	19.0	1.7	5.28
530	1	551.7	44.7	6.74	11.3	2.5	5.05
	5	695.0	22.9	6.84	8.7	1.2	4.94
	8	649.7	19.5	6.81	8.0	-	4.90
531	1	589.3	19.7	6.77	4.3	1.5	4.64
	5	600.3	39.2	6.78	3.0	-	4.48
	10	535.3	83.6	6.73	2.7	0.6	4.43
	14	556.7	21.5	6.75	3.3	1.2	4.52
532	1	264.7	39.6	6.42	<1.0	-	4.00
	5	580.3	96.0	6.76	<1.0	-	4.00
	10	213.7	43.9	6.33	<1.0	-	4.00
	20	2.0	-	4.30	<1.0	-	4.00
	35	93.0	-	5.97	1.7	0.6	4.22
533	1	327.3	71.4	6.52	<1.0	-	4.00
	5	299.7	49.7	6.48	<1.0	-	4.00
	10	81.7	17.4	5.91	<1.0	-	4.00
	20	2.3	0.6	4.37	<1.0	-	4.00
	42	50.0	13.9	5.70	<1.0	-	4.00
534	1	340.0	18.0	6.53	52.7	5.5	5.72
	5	1133.0		7.05	22.3	6.7	5.35
535	1	308.0	46.9	6.49	57.0	6.6	5.76
	5	1107.0	-	7.04	30.7	1.5	5.49
536	1	355.0	26.2	6.55	61.3	3.1	5.79
	5	773.7	11.1	6.89	34.0	8.7	5.53

Station Number	Depth (m)	ZoBell			PCA		
		Mean	S.D.	$Log_{10} x10^4$	Mean	S.D.	$Log_{10} x10^4$
537	1	254.3	20.8	6.40	82.0	3.6	5.91
	5	1040.0	-	7.02	79.0	13.0	5.90
	9	957.3	41.5	6.98	35.3	4.7	5.55
538	1	52.3	2.5	5.72	151.3	17.6	6.18
	5	194.7	28.9	6.29	148.3	11.2	6.17
539	1	49.7	5.7	5.70	114.7	21.7	6.06
	3	52.7	4.7	5.72	153.0	11.4	6.18
540	1	36.3	1.2	5.56	127.3	18.8	6.10
	3	984.0	26.0	6.99	-	-	-
541	1	863.3	98.2	6.94	70.0	5.0	5.84
	5	897.7	126.4	6.95	49.7	3.1	5.70
	10	480.3	22.0	6.68	83.7	9.3	5.92
	20	62.3	2.1	5.80	8.7	0.6	4.94
	33	50.7	13.6	5.70	9.0	2.0	4.95
542	1	1467.0	-	7.17	140.0	23.6	6.15
	5	916.3	73.1	6.96	98.3	9.5	5.99
	10	522.3	73.2	6.72	51.7	7.5	5.71
	20	25.0	2.0	5.40	4.7	0.6	4.67
	50	31.3	4.7	5.50	3.0	1.0	4.48
	75	31.3	4.7	5.50	1.7	0.6	4.22
	93	12.0	6.0	5.08	0.0	0.0	0.00